JOURNAL OF DAIRY SCIENCE

Contents

A Study of Cream Rising in Milk. F. M. SKELTON and H. H. SOMMER 321

Effect of Adding Cod Liver Oil to the Rations of Dairy Calves. T. W. GULLICKSON and J. B. FITCH 331

The Relation of Chlorine and Catalase Content of Milk to Its Curd Tension. E. G. HASTINGS and ARTHUR EREKSON 337

The Viscosity of Evaporated Milks of Different Solids Concentration. E. F. DEYSHER, B. H. WEBB and G. E. HOLM 345

The Bacteriology of Bull Semen. II. The Effect of Bacteria upon Rapid Tests for Semen Quality. I. C. GUNSALUS, J. J. R. CAMPBELL, G. H. BECK and G. W. SALISBURY 357

Peppergrass Silage. AVERY D. PRATT 365

A New Quantitative Method for Estimation of Total Combined Length of Mold Fragments in Butter. P. R. ELLIKER 369

The Determination of Citric Acid in Milk Products by Cerate Oxidimetry. BURDET HEINEMANN 377

The Effect of Iodinated Casein (Protamone) on Milk and Butterfat Production and on the Ascorbic Acid Content of the Milk. A. H. VAN LANDINGHAM, H. O. HENDERSON and CHARLES E. WEAKLEY, JR. 385

Program Thirty-Ninth Annual Meeting 397

Abstracts of Literature A75

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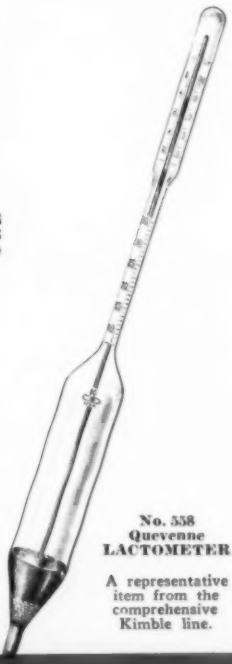

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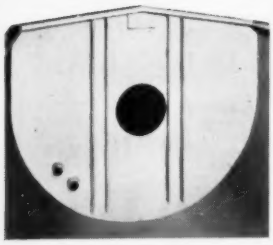

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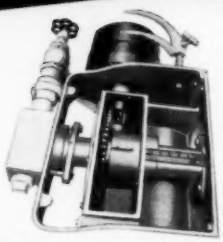

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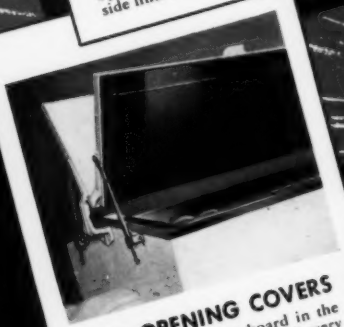

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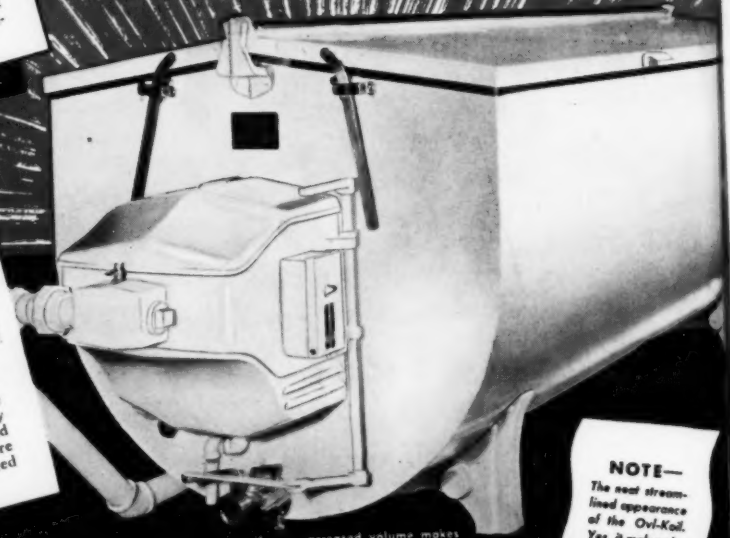

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JOURNAL OF DAIRY SCIENCE

VOLUME XXVII MAY, 1944 NUMBER 5

A STUDY OF CREAM RISING IN MILK

F. M. SKELTON AND H. H. SOMMER

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and

Department of Dairy Industry, College of Agriculture, University of Wisconsin, Madison, Wisconsin

The creaming process is a complex phenomenon that involves a number of factors. The difference in density between the fat and the milk plasma is the primary factor but a number of secondary factors, concerned with the formation of globule clusters, are required to make normal creaming possible. It has been shown that the individual globules rise in harmony with the velocity computed by Stokes' formula (4, 7, 8, 9), but this is too slow to account for the rapidity of normal creaming, the depth of the cream layer produced, and the sharp line of demarcation between the cream and skimmilk layers. The secondary factors, therefore, exert a controlling effect on the gravitational separation for creaming. The nature of these secondary factors and the detailed manner in which they function is still a matter of controversy.

The theory has been advanced that, aside from other factors, the gravitational movement of the globules (and clusters) is an important factor in cluster formation (1, 2, 5). The larger globules rising more rapidly overtake smaller globules, and as the clustering process proceeds in this manner, the clusters rise at an increasing rate and thus have increased opportunity for sweeping along individual globules as they occur in the upward path of the cluster. A difference in the fat content of the skimmilk at varied levels might well result from such a process. The study herein reported was undertaken to investigate this possibility.

EXPERIMENTAL

Samples of milk were placed in 1000-cc. graduated cylinders at 40° F. and stored in a refrigerator at that temperature. Six cylinders were used for each trial to provide for examination of a different cylinder at 1, 2, 4, 6, 10 and 24 hours after the cylinders were filled. Five experiments were conducted, one with fresh, raw milk and four with pasteurized milk. The

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milk had a fat content of 3.6 to 3.8 per cent. It was obtained from the mixed milk of about 40 herds as received commercially at the Department of Dairy Industry, University of Wisconsin. After the milk in the cylinders had stood undisturbed at 40° F. for the desired time, skimmilk samples were withdrawn at the following points as indicated by the graduated scale: 850, 750, 650, 550, 450, 350, 250, 150, 50 and 10 cc. The sample taken at the 850-cc. level was approximately 30 to 50 cc. below the cream line. The samples were withdrawn in the order as listed with the aid of the special apparatus shown in figure 1.

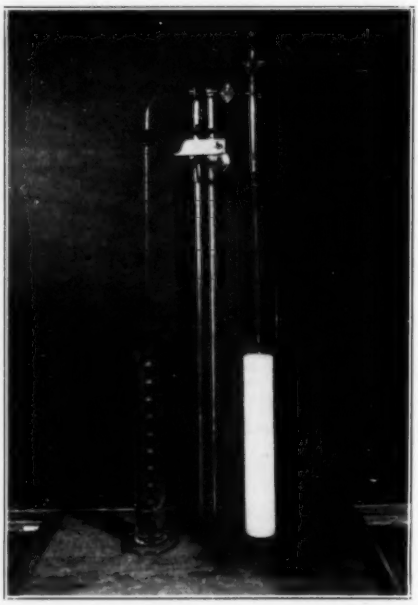

FIG. 1. Sampling device used for withdrawing samples of skimmilk at various levels from a graduated cylinder.

In the sampling procedure, entrance of the milk into the pipette was prevented until the tip was at the desired level. Mere closing of the mouth end of the pipette during lowering would not accomplish this since the hydrostatic pressure would compress the air and allow some milk to enter. This was counteracted to a point where a few small bubbles of air would actually escape from the pipette during immersion by simultaneously lowering a tube, connected to the pipette as shown in figure 1, into a sugar solution of sufficient density so that the hydrostatic pressure exceeded that caused by the milk. The assembly also carried a pointer, adjusted to the same level as the

pipette tip, to indicate its position as gauged by the pointer against the outside of the cylinder. When the pipette tip had been immersed to the desired level, the pipette was filled slowly with suitable manipulations of the valves and the application of suction. The entire assembly was then raised, the exterior of the pipette was wiped free from milk, and the sample within the pipette was then discharged into a suitable container. A clean, dry pipette was used for each sample. The fat content of the skimmilk samples was determined by the Mojonnier method as outlined by Mojonnier and Troy (3). All fat analyses were run in duplicate. The duplicate determinations

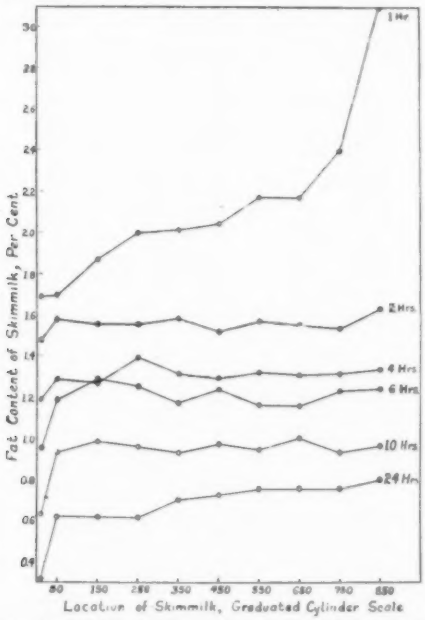

FIG. 2. Fat content of skimmilk at various levels below the cream line in 1000 cc. graduated cylinder used as a creaming vessel for whole pasteurized milk.

were made with considerable care and in general the difference between duplicates was very small in comparison with any observed differences in fat content between the various layers of skimmilk. Tables 1 to 5 give the duplicate fat tests and average fat tests of the skimmilk samples taken at various levels and at creaming times of 1 to 24 hours. The most rapid and extensive fat rising occurred during the first hour with a slow, gradual rise of the residual fat continuing during the succeeding 23 hours. While no data were secured beyond 24 hours, it can be assumed that a slow rising of residual fat would continue indefinitely in harmony with observations of Trelogan and Combs who studied rat rising in cream (6).

TABLE 1

Fat content of skimmilk secured below the cream line in raw milk at varied levels

Skim milk level	Average and duplicate fat determinations			
	1 hour		2 hours	
		Average		Average
850	2.749		1.218	
	2.743	2.746	1.219	1.218
750	2.222		1.193	
	2.216	2.219	1.205	1.199
650	2.051		1.157	
	2.052	2.051	1.161	1.159
550	1.910		1.179	
	1.910	1.910	1.179	1.179
450	1.783		1.179	
	1.782	1.782	1.176	1.178
350	1.736		1.214	
	1.746	1.741	1.218	1.216
250	1.708		1.243	
	1.711	1.709	1.244	1.244
150	1.603		1.210	
	1.615	1.609	1.210	1.210
50	1.555		1.175	
	1.552	1.554	1.165	1.170
10	1.500		1.118	
	1.499	1.500	1.122	1.120
	4 hours		6 hours	
		Average		Average
850	1.111		0.887	
	1.113	1.112	0.893	0.890
750	1.123		0.906	
	1.108	1.115	0.896	0.901
650	1.125		0.900	
	1.121	1.123	0.901	0.900
550	1.140		0.902	
	1.143	1.142	0.906	0.904
450	1.123		0.900	
	1.117	1.120	0.898	0.899
350	1.129		0.898	
	1.121	1.125	0.896	0.897
250	1.166		0.881	
	1.167	1.166	0.880	0.880
150	1.151		0.896	
	1.149	1.150	0.887	0.892
50	1.136		0.858	
	1.136	1.136	0.865	0.862
10	1.037		0.789	
	1.045	1.041	0.796	0.782

TABLE 2

Fat content of skimmilk secured below the cream line in pasteurized milk at varied levels

Skim milk level	Average and duplicate fat determinations					
	1 hour		2 hours		4 hours	
		Average		Average		Average
850	3.100		1.627		1.338	
	3.110	3.105	1.631	1.629	1.337	1.338
750	2.397		1.535		1.323	
	2.393	2.395	1.535	1.535	1.303	1.313
650	2.168		1.552		1.307	
	2.170	2.169	1.552	1.552	1.305	1.306
550	2.174		1.572		1.324	
	2.168	2.171	1.561	1.567	1.318	1.321
450	2.044		1.527		1.289	
	2.042	2.043	1.509	1.518	1.289	1.289
350	2.012		1.581		1.312	
	2.012	2.012	1.580	1.580	1.314	1.313
250	1.999		1.543		1.379	
	1.974	1.987	1.555	1.549	1.397	1.388
150	1.873		1.547		1.268	
	1.865	1.869	1.551	1.549	1.263	1.265
50	1.693		1.576		1.282	
	1.690	1.692	1.578	1.577	1.282	1.282
10	1.685		1.472		1.181	
	1.685	1.685	1.479	1.476	1.188	1.184
	6 hours		10 hours		24 hours	
		Average		Average		Average
850	1.240		0.966		0.802	
	1.242	1.241	0.961	0.964	0.801	0.802
750	1.227		0.934		0.757	
	1.227	1.227	0.935	0.935	0.757	0.757
650	1.162		0.997		0.755	
	1.153	1.158	1.001	0.999	0.751	0.753
550	1.161		0.944		0.752	
	1.160	1.160	0.943	0.944	0.754	0.753
450	1.236		0.969		0.730	
	1.237	1.237	0.972	0.970	0.720	0.725
350	1.179		0.932		0.702	
	1.163	1.171	0.930	0.931	0.698	0.700
250	1.252		0.959		0.613	
	1.250	1.251	0.958	0.958	0.613	0.613
150	1.284		0.984		0.613	
	1.283	1.284	0.983	0.984	0.617	0.615
50	1.180		0.931		0.620	
	1.185	1.182	0.932	0.932	0.617	0.618
10	0.956		0.628		0.321	
	0.952	0.954	0.628	0.628	0.307	0.314

TABLE 3

Fat content of skimmilk secured below the cream line in pasteurized milk at varied levels

Skim milk level	Average and duplicate fat determinations					
	1 hour		2 hours		4 hours	
		Average		Average		Average
850	2.711		1.709		1.368	
	2.705	2.708	1.696	1.703	1.362	1.365
750	2.420		1.696		1.293	
	2.408	2.414	1.697	1.697	1.305	1.299
650	2.325		1.525		1.292	
	2.327	2.326	1.533	1.529	1.280	1.286
550	2.225		1.540		1.277	
	2.215	2.220	1.560	1.550	1.257	1.267
450	1.997		1.515		1.255	
	1.986	1.992	1.524	1.520	1.253	1.254
350	1.969		1.527		1.246	
	1.971	1.970	1.531	1.529	1.240	1.243
250	1.925		1.562		1.327	
	1.925	1.925	1.551	1.557	1.330	1.328
150	1.839		1.531		1.277	
	1.852	1.846	1.531	1.531	1.276	1.277
50	1.823		1.510		1.337	
	1.815	1.819	1.506	1.508	1.322	1.330
10	1.620		1.399		0.994	
	1.615	1.618	1.410	1.404	0.994	0.994
	6 hours		10 hours		24 hours	
		Average		Average		Average
850	1.175		1.058		0.819	
	1.181	1.178	1.059	1.059	0.812	0.816
750	1.132		1.023		0.773	
	1.130	1.131	1.016	1.020	0.773	0.773
650	1.161		1.013		0.763	
	1.161	1.161	1.013	1.013	0.761	0.762
550	1.105		0.964		0.739	
	1.127	1.116	0.974	0.969	0.728	0.734
450	1.108		1.025		0.737	
	1.100	1.104	1.019	1.022	0.733	0.735
350	1.163		1.018		0.752	
	1.161	1.162	1.018	1.018	0.755	0.754
250	1.159		1.004		0.729	
	1.164	1.162	1.002	1.003	0.728	0.728
150	1.165		0.971		0.731	
	1.155	1.160	0.979	0.975	0.739	0.734
50	1.219		0.965		0.716	
	1.211	1.215	0.952	0.958	0.715	0.716
10	0.940		0.907		0.360	
	0.947	0.944	0.906	0.906	0.376	0.368

TABLE 4

Fat content of skimmilk secured below the cream line in pasteurized milk at varied levels

Skim milk level	Average and duplicate fat determinations					
	1 hour		2 hours		4 hours	
		Average		Average		Average
850	1.825		1.343		1.125	
	1.829	1.827	1.341	1.342	1.128	1.126
750	1.752		1.304		1.114	
	1.740	1.746	1.313	1.308	1.107	1.110
650	1.745		1.321		1.102	
	1.753	1.749	1.315	1.318	1.101	1.102
550	1.715		1.285		1.061	
	1.708	1.712	1.283	1.284	1.070	1.066
450	1.629		1.300		1.011	
	1.646	1.638	1.293	1.296.	1.064	1.062
350	1.620		1.318		1.045	
	1.618	1.619	1.331	1.324	1.052	1.048
250	1.360		1.283		1.062	
	1.576	1.568	1.291	1.287	1.069	1.066
150	1.501		1.260		1.077	
	1.499	1.500	1.271	1.266	1.083	1.080
50	1.438		1.233		1.080	
	1.444	1.441	1.236	1.234	1.070	1.075
10	1.347		1.170		0.988	
	1.352	1.350	1.182	1.176	0.981	0.984
	6 hours		10 hours		24 hours	
		Average		Average		Average
850	0.969		0.919		0.654	
	0.965	0.967	0.912	0.916	0.648	0.651
750	0.956		0.906		0.610	
	0.956	0.956	0.909	0.908	0.610	0.610
650	0.942		0.917		0.608	
	0.949	0.946	0.911	0.914	0.614	0.611
550	0.937		0.893		0.589	
	0.942	0.940	0.890	0.892	0.585	0.587
450	0.948		0.820		0.540	
	0.947	0.948	0.819	0.820	0.544	0.542
350	0.930		0.788		0.515	
	0.926	0.928	0.789	0.789	0.525	0.520
250	0.926		0.775		0.545	
	0.928	0.927	0.788	0.782	0.539	0.542
150	0.923		0.777		0.575	
	0.920	0.922	0.767	0.772	0.595	0.580
50	0.922		0.734		0.546	
	0.920	0.921	0.730	0.732	0.544	0.545
10	0.897		0.585		0.408	
	0.894	0.896	0.595	0.590	0.416	0.412

TABLE 5

Fat content of skimmilk secured below the cream line in pasteurized milk at varied levels

Skim milk level	Average and duplicate fat determinations					
	1 hour		2 hours		4 hours	
		Average		Average		Average
850	1.643		1.341		1.114	
	1.645	1.644	1.346	1.344	1.123	1.118
750	1.561		1.278		1.093	
	1.565	1.563	1.285	1.282	1.103	1.098
650	1.543		1.281		1.150	
	1.537	1.540	1.290	1.286	1.154	1.152
550	1.586		1.272		1.108	
	1.587	1.584	1.286	1.279	1.103	1.106
450	1.575		1.259		1.136	
	1.583	1.579	1.264	1.262	1.142	1.139
350	1.574		1.280		1.103	
	1.578	1.576	1.270	1.275	1.106	1.104
250	1.516		1.281		1.113	
	1.513	1.514	1.283	1.282	1.120	1.116
150	1.488		1.278		1.135	
	1.474	1.481	1.289	1.284	1.132	1.134
50	1.446		1.272		1.112	
	1.452	1.449	1.274	1.273	1.111	1.112
10	1.345		1.237		0.964	
	1.339	1.342	1.237	1.237	0.959	0.962
	6 hours		10 hours		24 hours	
		Average		Average		Average
850	0.950		0.929		0.603	
	0.946	0.948	0.935	0.932	0.602	0.602
750	0.967		0.860		0.657	
	0.963	0.965	0.856	0.858	0.608	0.662
650	0.968		0.855		0.599	
	0.972	0.970	0.862	0.858	0.600	0.600
550	0.964		0.871		0.608	
	0.964	0.964	0.877	0.874	0.618	0.613
450	0.973		0.878		0.621	
	0.971	0.972	0.874	0.876	0.617	0.619
350	0.989		0.867		0.610	
	0.980	0.984	0.874	0.870	0.616	0.613
250	1.006		0.870		0.592	
	1.008	1.007	0.867	0.868	0.604	0.598
150	0.967		0.890		0.587	
	0.960	0.964	0.897	0.894	0.590	0.588
50	0.966		0.865		0.601	
	0.966	0.966	0.861	0.863	0.607	0.604
10	0.870		0.598		0.467	
	0.868	0.869	0.595	0.596	0.462	0.464

In the gravitational theory of clustering, which inspired this study, it had been anticipated that, at some stages in the creaming process, the fat content of the skimmilk at some of the higher levels might actually be less than that of skimmilk at a lower point. This anticipation is on the basis of the greater sweeping action to which the fat globules in the upper portions of the skimmilk are subjected. The data indicate that such differences occur to a slight degree, but surprisingly the data suggest further that there is a wave-like progression of fat in the creaming process. This trend is revealed in the most pronounced manner by the data given in table 2 which have been presented graphically in figure 2.

No explanation is offered for the wave-like progression of the fat in the trials herein reported. While this tendency is suggested, the present approach is not ideal for a complete verification of this point. In order to establish this conclusion definitely, it would obviously be necessary to obtain a large number of points so that each wave in the graph might be defined by several points. It would then be necessary to use a method of sampling which would assure as far as possible a perfect horizontal section of the column of skimmilk at the selected level. While the present equipment was intended to attain a sample of skimmilk at a precise level, its use in sampling at still closer intervals probably would not be justified because of the manner in which the milk flows into the pipette.

SUMMARY

It was shown that in the creaming of milk the greater portion of the fat rises during the first hour. A slow, gradual rise of residual fat continues indefinitely. There is some indication in the data that during the creaming process the fat content of the skimmilk at one level may be slightly less than the fat content of the skimmilk at a lower level. Indications were obtained that such differences may occur at several points in a skimmilk column suggesting a wave-like progression of the fat. This suggestion should be further verified before it is accepted as a definite conclusion. There appears to be no difference in the manner in which the cream layer forms on raw milk as compared with pasteurized milk.

REFERENCES

(1) BANCROFT, W. D. Applied Colloid Chemistry. McGraw-Hill Book Co., New York. 1921.

(2) FLEISCHMANN, W. Lehrbuck der Milchwirtschaft. Bremen. 1893.

(3) MOJONNIER, T., AND TROY, H. C. The Technical Control of Dairy Products. Mojonnier Bros. Co., Chicago, Ill.

(4) RAHN, O. Molkerei Zeitung (Hildesheim), **38**: 1321. 1924.

(5) SOMMER, H. H. Market Milk and Related Products, pp. 646–649. Published by the author, Madison, Wis.

(6) TRELOGAN, H. C., AND COMBS, W. B. Fat Rising in Cream. JOUR. DAIRY SCI., **17**: 675–684. 1934.

(7) TROY, A. C., AND SHARP, P. F. Data published in "Fundamentals of Dairy Science," Chemical Catalog Co. 1928.
(8) VAN DER BURG, B. Forsch. Geb. Milchwirt-schaft. Molkereiwesen, 1: 154. 1921.
(9) VAN DAM, W., AND SIRKS, H. A. Verslag. Land. Onderzoek. Rijkslandbouwproefsta., 26: 106. 1922.

EFFECT OF ADDING COD LIVER OIL TO THE RATIONS OF DAIRY CALVES*

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Some calves dropped in the dairy herd at University Farm are weak at birth, and develop indigestion or other ailments and a few die before they are a month old. The cows in the herd are fed largely on a barn-feeding basis throughout the entire year because little or no pasture is available. Fair to good quality roughage is fed. Meigs and Converse (6) have shown that the quality of roughage fed to cows has a marked effect on the health and vigor of their calves at birth. Similarly the beneficial effect of feeding a vitamin A supplement to calves for several weeks after birth or until they are consuming considerable quantities of hay has been demonstrated by Phillips (7). If carotene-poor roughage is fed, or if young calves, for any reason, fail to utilize the carotene supplied then the feeding of a vitamin A-rich product beyond the first few weeks should prove beneficial. Ellenberger (4) reported that neither calves raised to 240 days of age on poor quality roughage nor those fed good hay for the same period evidenced any lack of vitamin A. Calves fed the poor hay, however, made "slower, lesser, and more costly growth" as caused by their lower consumption of nutrients in the less palatable ration. When good quality hay was fed, no benefits appeared from feeding cod liver oil, instead growth in some cases seemed to be retarded. Cary (2) states that calves fed skim milk, grain and hay after 30 to 60 days of age must, in order to survive, receive a vitamin A supplement if fed hay as poor in carotene as U. S. No. 3 alfalfa or timothy. Feeding at the rate of 20 milliliters of cod liver oil per calf daily to six months of age, however, had no demonstrable effect on their general appearance and rate of growth when U. S. No. 2 alfalfa hay or better was fed. Similarly, Insko and Rupel (5) observed no marked difference in the rate of growth and composition of bones between calves fed a standard ration, including good quality legume hay, and others reared on a similar ration supplemented with cod liver oil. Similar results have more recently been reported by Reaves and Cannon (8). In a camparison involving four pairs of calves from dams that presumably obtained a normal allowance of pasture, Dahlberg and Maynard (3) fed varying amounts of Nopco XX, a vitamin A and D rich concentrate of cod liver oil. Both lots weighed exactly the same at the start, but the supplemented group soon forged ahead and at 26 weeks of age each averaged 18.7 pounds more than those in the check group. The

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authors concluded that if there was any difference in general health it was in favor of the calves receiving the supplement.

EXPERIMENTAL

The object of the experiment reported here was to determine the effect, if any, on the growth and well-being of dairy heifer calves at University Farm of supplementing their rations with cod liver oil from birth to six

TABLE 1

Effect on the weight of calves of various breeds of including cod liver oil in their rations to 180 days of age. Data calculated to a 10-day interval basis

Age	Guernseys		Holsteins		Jerseys	
	Non-cod liver oil	Cod liver oil	Non-cod liver oil	Cod liver oil	Non-cod liver oil	Cod liver oil
	No. in group					
	7	12	14	14	12	13
days	*pounds*	*pounds*	*pounds*	*pounds*	*pounds*	*pounds*
Birth	64.0	65.9	94.7	93.6	53.9	53.2
10	66.4	67.2	101.2	97.9	55.8	56.5
20	70.6	72.4	112.7	108.9	62.2	63.3
30	80.5	80.2	126.1	121.8	68.5	72.1
40	86.4	88.3	137.0	138.4	76.8	82.4
50	97.5	94.6	152.6	152.6	90.4	90.0
60	104.4	102.2	171.0	169.6	101.6	101.2
70	114.6	114.4	187.1	185.0	112.6	113.8
80	126.7	127.8	201.8	201.7	122.4	127.2
90	136.1	142.6	219.7	217.3	134.1	141.5
100	149.6	157.4	235.7	235.2	148.5	157.4
110	162.7	171.9	256.0	251.9	159.3	170.2
120	174.9	188.9	274.8	271.8	173.7	186.0
130	189.4	204.5	292.6	290.4	185.5	197.0
140	204.6	222.5	309.5	307.5	201.5	211.2
150	217.2	239.3	329.1	326.2	216.3	226.6
160	235.5	255.1	347.6	350.2	232.9	244.2
170	250.8	270.8	367.3	370.0	247.3	261.1
180	262.5	286.9	389.1	389.0	258.7	275.5
Total gain in 180 days	198.5	221.0	294.4	295.4	204.8	222.3
Average daily gain	1.10	1.23	1.64	1.64	1.14	1.24

months of age. The trials were carried on with all the heifer calves born in the herd during a period of about three years. It included 19 Guernseys, 25 Jerseys and 28 Holsteins. The calves were assigned at birth alternately to the check and experimental groups. The calves in both groups were fed and handled alike except that 25 to 35 cc. of cod liver oil (U.S.P.) was fed daily to each animal in the supplemented group. Whole milk was fed the first 30 days, and then skim milk to six months of age. The milk was fed at the rate of one-eighth the weight of the animal but not over 16 pounds daily per calf. Calves were allowed to eat all the alfalfa hay they wanted

at all times. Fair to good quality hay was fed. A grain mixture consisting of equal parts by weight of oats, corn, and barley (all ground medium fine), and wheat bran was fed *ad libitum* up to four pounds daily per calf. Water and salt was available to the calves at all times. Calves were kept in large roomy pens with four or five of the same breed and of approximately the same age in each. They were allowed to exercise outdoors several hours daily when weather conditions permitted.

Each calf was weighed within a few hours after birth and at regular ten-day intervals thereafter. Measurements of height at withers were made every 30 days beginning within a few weeks after birth. Frequent observations were made of the physical condition and appearance of each calf.

TABLE 2

Effect on the height at withers of calves of various breeds of including cod liver oil in their rations to 180 days of age. Data calculated to a 30-day interval basis

Age	Guernseys		Holsteins		Jerseys	
	Non-cod liver oil	Cod liver oil	Non-cod liver oil	Cod liver oil	Non-cod liver oil	Cod liver oil
	No. in group					
	7	12	14	14	12	13
days	*cm.*	*cm.*	*cm.*	*cm.*	*cm.*	*cm.*
10	70.4	71.8	74.8	74.1	67.3	67.9
30	72.4	73.5	77.6	78.1	69.4	69.8
60	76.6	76.8	83.4	83.6	75.1	75.0
90	81.2	81.2	89.4	88.8	80.1	80.8
120	86.0	86.6	94.2	94.0	84.8	86.3
150	90.4	91.8	99.2	98.6	89.8	90.5
180	94.7	96.2	104.2	103.1	93.4	95.0
Total gain in height in 170 days	24.3	24.4	29.4	29.0	26.1	27.1
Average daily gain	0.14	0.14	0.17	0.17	0.15	0.16

RESULTS AND DISCUSSION

Data are presented in table 1, showing the weights at ten-day intervals, from birth to 180 days of age, of groups of calves of the Guernsey, Holstein, and Jersey breeds respectively, when fed cod liver oil and of corresponding non-supplemented groups. Data relating to the average height at withers of the animals in the various groups at 30-day intervals are presented in table 2. As height measurements of calves at birth were seldom made, none are given in the table.

From the data in tables 1 and 2 it is apparent that adding cod liver oil to the rations had no evident effect on the rate of gain in height at withers of calves of any breed nor on the gain in weight of Holsteins. On the other hand the Guernseys and the Jerseys fed cod liver oil, gained an average of

22.5 pounds and 17.5 pounds more respectively per calf during the 180 days than those in corresponding non-supplemented groups. Statistical analysis of the data indicate that these differences are slightly significant (9).

The recorded observations indicate that less trouble from digestive disturbance was encountered in young calves that were fed cod liver oil than among those in the non-supplemented groups. It is also significant that a few calves in the non-cod liver oil groups died within a few weeks after birth, and others apparently were saved from this fate when cod liver oil was added to their rations. These facts largely account for the difference in number of calves in the supplemented and the non-supplemented groups. Data relating to the calves that died during the experimental period or that were transferred to a supplemented ration are not included in tables 1 and 2.

Contrary to what may have been expected, as based on the studies of Phillips (6) the cod liver oil supplement had no apparent effect on the rate of gain of calves until after they were about 90 days old. It seems probable that the whole milk, which, generally is a rich source of vitamin A and carotene, helped to build up a small reserve of these factors in the body of the calf during the first 30 days when it was fed and this together with the carotene obtained in the limited quantity of hay consumed provided sufficient vitamin A for growth to about 90 days of age. Beyond that age, however, with the vitamin A reserve of the body largely exhausted the supply obtained from the roughage in the ration was not fully adequate except in the case of the Holstein calves. This difference may have been due to a higher carotene requirement by the Guernsey and the Jersey calves as has been suggested by Boyer *et al.* (1), or to their inability to utilize this factor as efficiently as the Holsteins. Also, it is possible that the Holstein calves consumed proportionately more roughage and thus obtained correspondingly more carotene. Unfortunately complete records were not kept of the quantity of hay consumed by each calf. To what extent results would have been changed had excellent, instead of fair to good, quality hay been fed can only be surmised. From the data presented it appears that it may be beneficial to feed cod liver oil to young calves, especially Guernseys and Jerseys, when roughage only fair in quality is fed.

SUMMARY

Comparisons were made using a total of 72 calves, including 19 Guernseys, 28 Holsteins and 25 Jerseys, to determine the effect, if any, of adding cod liver oil, as a vitamin A supplement to the rations commonly fed to calves during the period to six months of age. Alternate animals were fed 25–35 cc. cod liver oil daily beginning when the calf was only a few days old, in all other respects the plan of feeding was alike for both groups.

Somewhat less digestive troubles occurred in the calves fed cod liver oil than among those in the non-supplemented groups. No significant differ-

ence was noted in any breed in rate of gain in height at withers between the calves fed cod liver oil and those in the check group. Adding cod liver oil to the ration had no evident effect on the rate of gain in weight of Holstein calves. The Guernsey and the Jersey calves fed cod liver oil, on the other hand, gained an average of 22.5 pounds and 17.5 pounds more per calf respectively during the 180-day period than those fed similar but non-supplemented rations.

REFERENCES

(1) Boyer, P. D., Phillips, P. H., Lundquist, N. S., Jensen, C. W., and Rupel, I. W. Vitamin A and Carotene Requirements for the Maintenance of Adequate Blood Plasma Vitamin A in the Dairy Calf. Jour. Dairy Sci., 25: 433–440. 1942.

(2) Cary, C. A. The Vitamin A Requirements of Very Young Calves. Report of the Chief of the Bureau of Dairy Industry, U. S. Dept. Agr., pages 34–36. 1941.

(3) Dahlberg, A. C., and Maynard, L. A. Note on Feeding Vitamin A and D Concentrate in Cod Liver Oil to Calves. Jour. Dairy Sci., 20: 59–62. 1937.

(4) Ellenberger, H. B. Effect of Feeding Vitamin A and D in Concentrated Cod Liver Oil with Different Grades of Hay to Dairy Calves. Vt. Agr. Expt. Sta. Bul. 425: 23–24. 1937.

(5) Insko, W. M., and Rupel, I. W. Cod Liver Oil Fails to Improve a Good Dairy Calf Ration. Wis. Agr. Expt. Sta. Bul. 405: 50. 1929.

(6) Meigs, E. B., and Converse, H. T. Physiological Effects of Rations with Low Grade Roughage. Abst. Papers Presented at the 27th Ann. Meeting, Amer. Dairy Sci. Assoc., page 55. 1932.

(7) Phillips, Paul H., Lundquist, Norman S., and Boyer, Paul D. The Effect of Vitamin A and Certain Members of the B-complex upon Calf Scours. Jour. Dairy Sci., 24: 977–982. 1941.

(8) Reaves, P. M., and Cannon, C. Y. Supplementing a Normal Ration for Dairy Calves with Cod Liver Oil. Jour. Dairy Sci., 19: 436. 1936.

(9) Snedecor, George W. Statistical Methods. The Iowa State College Press, Ames, Iowa. 67–78. 1940.

THE RELATION OF CHLORINE AND CATALASE CONTENT OF MILK TO ITS CURD TENSION*

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The type of curd obtained from milk with rennet has been studied by many in attempts to relate the nature of the curd to one or more of the properties of the milk. The reaction of the enzyme and the milk will vary from sample to sample in its speed, that is, in the time required for the rennet to cause visible curdling under standard conditions of temperature and concentration of enzyme. The physical properties of the coagulum will also vary from sample to sample. Most observations have been limited to the product of individual cows and also to relatively short periods. A field project on the manufacture of cheese presented an opportunity to study curdling properties and milk composition of each of six herds daily throughout a lactation period and the milk of each animal in each herd several times in the period.

The animals of the six herds, totaling 157 milking cows, freshened in early spring and were dried off in early November. The resting time between lactation periods was thus longer than in herds in which production is maintained at a relatively constant level throughout the year.

Firmness of the curd produced under standard conditions of temperature and concentration of rennet, chlorine and catalase content of the sample, and also the reducing power of each sample toward methylene blue and toward resazurin were observed. The accumulated experience of many has shown that the mass of reproducing bacteria of the milk is the chief reducing constituent of milk toward methylene blue and that the mass of growing bacteria and the mass of body cells in milk are the reducing agents toward resazurin. The action of each of these two agents on resazurin can be separately determined in some degree when the reducing power of the milk on this reagent is compared with the action on methylene blue. The reduction of resazurin can be divided into two phases: first, a change from the original slate gray color of the milk-resazurin mixture to a pink, and second, a change of the latter to white. The extent of the progress in each phase can be observed. If the content of a milk in body cells (leucocytes) or bacteria is high, the time required for the first phase of the reduction of resazurin will be short. The answer as to which of the reducing agents is responsible for a color change noted with resazurin is supplied by comparing results of the two

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tests. If the bacteria are the chief cause of reduction in any sample, reduction times with the two dyes will be much the same. If, however, the methylene blue indicates presence of few bacteria, then the rapid reduction of resazurin to its pink form is due to body cells, the presence of which is indicated by the high catalase content, since the body cells are apparently the source of the catalase of milk. Its other constituents are devoid of this enzyme. The chlorine content of milk high in catalase will also be high since blood serum will also be entering the milk system if cells are passing from the blood system into the milk system. Thus chlorine, catalase and body cells will tend to vary together in the milk of an individual cow and these variations will cause variations in reducing power of the milk toward resazurin. High bacterial content will tend to obscure the latter relation.

The entrance of blood serum into the milk system tends to raise the pH of the milk and increase the sodium: calcium ratio, changes that alter the

TABLE 1

The season's history of the herds studied

Herd	May 9				Sept. 30				Oct. 24			
	Cows		Milk		Cows		Milk		Cows		Milk	
	No.	%	*lbs.*	%	*No.*	%	*lbs.*	%	*No.*	%	*lbs.*	%
1	34	100	1120	100	24	70	576	60	20	60	364	32
2	33	100	902	100	31	94	446	49	31	91	324	36
3	27	100	791	100	27	100	569	72	25	92	387	49
4	21	100	450	100	17	81	235	52	14	66	242	53
5	24	100	576	100	21	88	497	86	21	88	314	54
6	18	100	606	100	17	94	360	57	15	83	258	42
Average	157	100	741	100	137	87	447	60	126	80	315	42

speed of reaction of milk with rennet and also the properties of the coagulum. It seems probable that the presence of an inflammatory process in the udder causes a decrease in the percentage of casein from that normal for the animal and changes its quality in such a manner that curdling with rennet is retarded and the firmness of the curd is decreased from that normal for the animal. It is also to be remembered that the curds formed in the milk of different cows by rennet will vary in physical properties due to the genetic pattern of the animal. In this case the four quarters of the udder should produce milk showing the same type of coagulum, while in the case of an inflammatory process the nature of the coagulum noted in the product of the affected quarter or quarters would differ from that of the normal quarter.

This study is one of the relation of mastitis to the quality of milk and to the quality of the cheese made therefrom. The discussion is based wholly on comparative values, so it does not seem essential to describe in detail the methods used. The property of the curd observed was firmness or tension.

The seasonal record of the herds is presented in table 1.

The curd tension of the entire quantity of milk produced in 24 hours was noted once or twice each week throughout the season. Readings were made 30 and 40 minutes after the rennet was added to the milk. The relative values were the same in either case. Only those noted after 30 minutes are given in table 2. The product of herd 4 is markedly different in curd tension from that of the other herds.

In the detailed study of each herd it seemed that the clearest picture of the relation of mastitis to curd tension and to other items of interest would be obtained by comparing animals that in May produced milk of a curd tension of 5 or below with animals that during the same period produced milk having a curd tension of 40 or above. One point of interest is the relation of curd tension to persistence in production. The data are presented in table 3. The significance of the data is only indicative on account of the limited number of animals in each herd.

TABLE 2

Average curd tension by months of herd milk

Herd	1	2	3	4	5	6
May	13	17	18	22	12	14
June	9	9	6	13	3	4
July	10	12	5	14	4	8
August	4	6	5	11	2	4
September	5	7	4	16	2	5
October	12	14	12	20	10	9
Average	8.8	10.8	8.3	16.0	5.5	7.3

The entire comparison may be thus expressed. Twenty-five cows that produced milk of a curd tension of 5 or below in May gave in late September 53.1 per cent as much milk as in May. Twenty-four cows that in May produced milk with a curd tension of 40 or above gave in September 65.0 per cent as much milk as in May. The observations conform with what one would expect in showing that mastitis is a factor in persistence of production in any lactation period.

CURD TENSION IN RELATION TO CHLORINE AND CATALASE CONTENT

The relationship of curd tension to chlorine and catalase content of milk was also studied, using only the extreme cases, *i.e.*, those with a curd tension of 5 or below and those with a curd tension of 40 or above. The judgment as to the members of the intermediate group would be questionable, hence the use of the extremes rather than the entire number of cows in the six herds.

The catalase content of the milks was determined by collecting the total amount of oxygen set free from an excess of hydrogen peroxide by a quantity of the milk and expressing it in terms of percentage of the quantity of milk

TABLE 3

The relationship of curd tension to persistence in production

	Curd tension	On Sept. 25 produced of the May 8 yield
Herd 1		*Per cent*
3 cows	5 or less	24.7
3 cows	40 or more	68.0
Herd 2		
9 cows	5 or less	42
3 cows	40 or more	50.0
Herd 3		
5 cows	5 or less	74
3 cows	40 or more	60
Herd 4		
0 cows	5 or less	.....
6 cows	40 or more	65
Herd 5		
7 cows	5 or less	92
2 cows	40 or more	100
Herd 6		
1 cow	5 or less	33
7 cows	40 or more	48

used, thus if 10 cc. of milk set free 10 cc. of oxygen its catalase value would be 100.

The values given in table 4 are the average of all observations during the entire period of observation. The relation of high chlorine and catalase content to low curd tension is to be noted in each herd except herd 5. Since the record of this herd differs so markedly from that of the other five herds, the detailed data are presented in table 5. Herd 5 contained many cows producing milk of low curd tension with no evidence of udder trouble as shown by the chlorine and catalase content of the milk. It has long been known that some cows normally produce soft-curd milk, while in others the changed composition of the milk is due to a disturbance in the physiology of the

TABLE 4

The catalase and chlorine content of herd milk of low and high curd tension in May

Herd	Curd tension			
	5 or less		40 or more	
	Chlorine	Catalase	Chlorine	Catalase
	%	%	%	%
1	0.169	145	0.117	34
2	0.170	97	0.125	38
3	0.146	52	0.128	15
4	.....	.....	0.117	26
5	0.114	18	0.130	10
6	0.184	59	0.121	22

udder. As far as the writers know, this is the first instance of a herd of apparently normal cows producing milk of low curd tension or soft-curd milk. The herds studied consist largely, if not wholly, of their own progeny; the genetic factor responsible for soft-curd milk may thus be brought to attention in a degree not probable in a herd maintained by purchase.

TABLE 5

The catalase and chlorine content of the milk of individual cows of low and high curd tension in herd 5

Cow	Curd tension	Chlorine	Catalase	Cow	Curd tension	Chlorine	Catalase
			May 9, 1941				
23	0	0.114	17	1	63	0.135	10
				5	48	0.122	14
				12	42	0.133	6
			May 26, 1941				
6	0	0.085	7	2	48	0.088	2
13	0	0.102	15				
14	0	0.091	4				
15	0	0.097	28				
18	0	0.091	4				
19	0	0.093	17				
22	0	0.101	5				
23	0	0.083	10				
25	0	0.092	6				
27	0	0.089	35				
			July 21, 1941				
6	0	0.082	12	1	42	0.087	17
7	0	0.100	3	2	52	0.082	2
14	0	0.102	6	4	65	0.101	—
15	0	0.079	50	5	43	0.086	5
18	0	0.103	10	11	40	0.098	14
22	2	0.103	9				
23	0	0.099	7				
27	5	.....	18				
				17	44	0.103	9
				24	43	0.103	33
			Aug. 7, 1941				
6	0	0.079	21	2	65	0.081	5
7	0	0.066	10	12	52	0.062	14
14	0	0.086	18	17	50	0.097	13
15	0	0.100	69	24	42	0.106	36
18	0	0.107	17				
22	0	0.087	39				
23	0	0.084	30				
27	4	0.090	47				

The resazurin tests were observed at the end of each hour for five hours, and a number given to each sample at each observation. A low number, 1, for example, indicates that little change in the color of the dye was noticed

TABLE 6

The relationship of resazurin reduction after 1 hour incubation to curd tension, chlorine and catalase content of milk

Herd	Curd tension	Cows	Chlorine	Catalase	Resazurin
1	5 or less	6	0.169	145	4.0
1	40 or more	10	0.117	34	2.0
2	5 or less	15	0.170	97	3.5
2	40 or more	3	0.125	38	1.0
3	5 or less	6	0.146	52	1.6
3	40 or more	3	0.128	15	1.0
4	5 or less	0	.....	.....	.....
4	40 or more	7	0.117	26	1.5
5	5 or less	1	0.114	17	1.0
5	40 or more	3	0.130	10	1.3
6	5 or less	2	0.184	59	4.0
6	40 or more	6	0.121	22	1.5

at that period of observation, while a high number indicated much greater reduction at the period of observation. Thus, 1 at the fourth hour means a milk very low in bacterial content and in cells, while 5 at the first hour means a milk high in bacteria or in body cells, or in both. The answer as to which is given by the methylene blue test. The milks in question were constantly so low in bacteria that a rapid reduction with resazurin may be used as an evidence of high content in body cells. It thus seems proper to use the averages of the resazurin reading of the two classes of cows—those producing milk of a curd tension of 5 or below and those showing a tension of 40 or above.

Table 6 presents the average reading of the resazurin test at the end of the first hour for the two groups of cows in each of the six herds on a particular day in May. The expected relation between high chlorine and catalase content and rapid reduction of resazurin is shown in each herd except 5, in which the more rapid reduction was noted in the high tension group of cows. The number of cows in each group of this herd was so small that the figures can be only suggestive. The smallness of the groups representing the extreme high and low values indicates that the herd consists largely of normal animals.

TABLE 7

The relationship of catalase content of milk to the rapidity of reduction of resazurin

Herd	Average catalase content	Average reading in resazurin test, 1 hr.
1	56	3.0
2	65	3.2
3	48	2.5
4	29	1.9
5	25	1.4
6	48	2.7

The relationship between the average catalase content of the entire amount of milk delivered at the factory to the average reading of the resazurin test at the end of the first hour for each herd is shown in table 7.

The relationship between catalase content and reducing power toward resazurin is clearly evident from the data, which also show that herd 5 is the only one of the six that can be considered as producing normal milk, since the milk of cows with healthy udders should not exceed 25 in its catalase value. Indeed, the value 20 is most frequently found in the literature on the subject, and in the experience of the writers some herds will consistently produce milk that will not exceed this value. The same is true of most first lactation cows.

The cause of the condition noted in herds 1, 2, 3 and 6 cannot be stated with certainty. From the few observations made as to the presence of streptococci in the milk of individual cows, one is inclined to consider them as the prime disturbing agent. The observations of the senior author indicate that herds in similar condition are to be found in all areas. It is hoped that the incidence of such herds is not as high as in the six studied, for they cannot serve as the foundation of a successful farm or of a successful cheese industry. Some program of herd management should be developed through the use of which such herds can be placed on a more normal base.

SUMMARY

The tension of the curd produced under standard conditions in the 24-hour product of each of six herds was noted frequently throughout a period of six months. The product of one herd (No. 4) had an average curd tension approximately twice as great as that of the other five herds and is believed to represent what one would expect to obtain from a group of cows with healthy udders. The low curd tension of four of the remaining five herds is believed to be due to the high incidence of chronic mastitis in these herds, since the product thereof was abnormally high in catalase and in chlorine. The low curd tension noted in the product of the remaining herd (No. 5) is believed due to the genetic pattern of the members of the herd since the chlorine and catalase content of its product was normal.

The record of the four herds indicates them to be agents of low value for the production of milk both from the standpoint of quantity and quality of milk.

THE VISCOSITY OF EVAPORATED MILKS OF DIFFERENT SOLIDS CONCENTRATION

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The viscosity of evaporated milk is an important characteristic because, in the minds of many consumers, it indicates a quality of "richness." Of greater significance, however, is the fact that an increase in the viscosity of the evaporated milk retards the rate of fat-separation in storage, since the phenomenon of fat-globule-rise is largely a function of size of globules and viscosity of medium. Objectionable fat-separation may occur because of improper homogenization of the milk or because the body of the evaporated product is too "thin." This report is a discussion of a preliminary study of the viscosity of evaporated milks of different concentrations and of the effects of different manufacturing and storage conditions.

Some years ago preliminary observations were made in these laboratories on evaporated milks that contained approximately 26 per cent of milk solids. Forewarming the whole milks up to 100° C. (212° F.) was carried out in a steam-jacketed hotwell; forewarming at 100° C. to 120° C. (248° F.) was accomplished in an autoclave. The reciprocal relationship of heat stability and consistency was noted in many instances. For example, forewarming a milk at 70° C. (158° F.) for 10 minutes usually rendered its evaporated product less stable than an evaporated product from a milk heated to 95° C. (203° F.) for 10 minutes. However, as a rule the consistency of the former product at coagulation was greater. In several cases, milks heated to 105° C. (221° F.) for 10 minutes produced evaporated products of greater stability than those from other samples forewarmed at lower temperatures. The consistencies of the milks heated to the higher temperatures were less.

These results were similar to those obtained by Grindrod (1), who subjected milk to the "impact process" of sterilization and found that milk heated to 110° C. for one minute possessed greater heat stability than did milk forewarmed at the commercially used temperatures (90°–100° C.). The resulting evaporated milk was too fluid.

Hunziker (2) states that forewarming temperatures above the boiling point produce maximum stability but low viscosity while the reverse is true at lower forewarming temperatures. Recent work in these laboratories (5, 6) has shown that the heat stability and concentration of evaporated milks can be greatly increased by the use of high temperatures applied to the milk for a short time. By means of this method of treatment, samples with a wide range of concentrations and stabilities were made available for viscosity determinations.

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EXPERIMENTAL

Whole milk from the Beltsville herd of the Bureau of Dairy Industry was standardized to a fat to solids-not-fat ratio of 1 to 2.29. Experimental samples were prepared, both with the usual forewarming and with the high-temperature forewarming procedure described elsewhere (5, 6). The milks forewarmed at 95° C. for 10 minutes were heated in a steam-jacketed hotwell. Those forewarmed by the high-temperature short-time method were heated in a Mallory heater (3).

After the milk was forewarmed it was concentrated in a stainless steel vacuum pan to 32–35 per cent solids, heated to 60° C., homogenized at a pressure of 2500 pounds per square inch, and cooled. Precautions were taken throughout the work to obtain uniform homogenization in all batches and thus to eliminate any variable which might result from a lack of uniformity in fat-globule size. Small quantities of the milk were diluted with water to the desired solids content. The heat stability, or time required to initiate coagulation, in these samples at 115° C. was then determined by heating them in small cans in a pilot sterilizer. The sterilizer reel revolved at the rate of 4 rpm. and during the treatment of all samples it was allowed to run continuously.

Viscosity determinations were made on 150-gram samples with a McMichael viscosimeter. The measurements were made at 20° C., unless otherwise stated. The wires were standardized against sugar solutions of known viscosity and all results were converted to centipoises. The cans of milk were stored in constant-temperature rooms, undisturbed, until viscosity determinations were to be made. Samples were adjusted to the temperature of measurement by immersing the cans in a water bath for several hours before opening them. Excessive fat-separation in occasional samples made it impossible to obtain accurate measurements on such milks, and consequently some inconsistencies in the data were caused by this change in fat-dispersion.

RESULTS

The data plotted in figures 1 and 2 show the course of viscosity development in concentrated milks of different types during heating to and beyond the point of coagulation. The effect of aging milk and of the addition of lactic acid upon the body produced during sterilization is shown in figure 1. The curves in figure 2 show the viscosities of milks of different concentrations after various periods of heating at 115° C. The samples of figure 1 were forewarmed by usual commercial methods (95° C. for 10 minutes) while those of figure 2 (except the "95°" controls) were forewarmed at high temperatures and held for 25 seconds. These figures indicate how rapidly the viscosity of the evaporated milks increased just before and after coagulation. The viscosities at coagulation decreased as the heat stabilities of the milks increased.

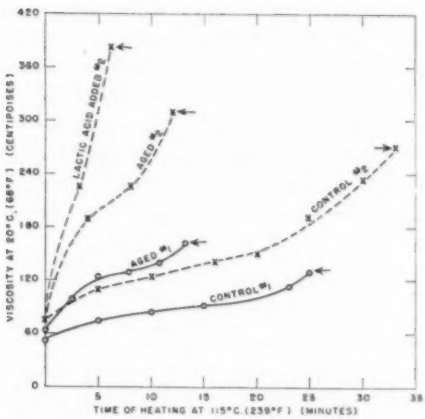

FIG. 1. The relationship between the heat stability of concentrated milk and the viscosity which is developed in it during heating at 115° C. (239° F.). The arrows indicate the time of the appearance of visible coagulation. All milks were forewarmed at 95° C. (203° F.) for 10 minutes before concentration to 26 per cent solids content.

The relationship between the viscosities of evaporated milks of different solids content and their heat stabilities is shown in figure 3. The body of the

TABLE 1

The effect of differences in heat stability upon the viscosity of evaporated milks sterilized at 115° C. (239° F.) for 18 minutes and held in storage. The different stabilizing heat treatments used on the raw and concentrated milks are indicated. Samples bearing the same date were prepared from a common sample of raw milk.

Date	Pre-sterilization heat treatment of milk				Total solids	Heat stability	Viscosity at 30° C. (86° F.) after storage at 30° C.				
	Raw milk		Concentrated milk				No storage	10 days	30 days	225 days	410 days
	Temp.	Time	Temp.	Time							
	°C.	*min.*	*°C.*	*min.*	*%*	*min.*	*c.p.*	*c.p.*	*c.p.*	*c.p.*	*c.p.*
12/2/41	95	10	.....	.....	26.0	44	26	12	13	13	.....
12/29/41	140	1	.....	.....	31.7	23	174	95	47	51	.....
2/13/42	95	10	.....	.....	26.0	43	30	19	16	24	.....
	120	2	.....	.....	34.9	25	460	222	156	360	.....
6/25/42	95	10	.....	.....	27.7	23	58	24	19	18	15
	95	10	120	0	27.7	34	25	16	15	14	15
	95	10	130	0	32.5	44	66	43	37	53	55
	95	10	140	0	37.2	35	165	132	121	213	191
7/9/42	95	10	120	4	35.0	22	207	165	170	197	290
7/13/42	95	10	.....	.....	27.6	29	45	18	16	15	17
	95	10	120	0	27.6	46	24	14	13	18	20
	120	1/4	.....	.....	27.6	36	34	14	13	16	49

milk was greatly influenced by the heat stability of the sample while one of the important factors affecting stability was concentration. The effect of heat stability upon the viscosity of some milks of different concentrations is shown also in table 1.

Results which show the extent of the viscosity changes in evaporated milk subjected to different manufacturing and storage conditions are presented in figures 4 and 5 and in table 1. The important changes during storage

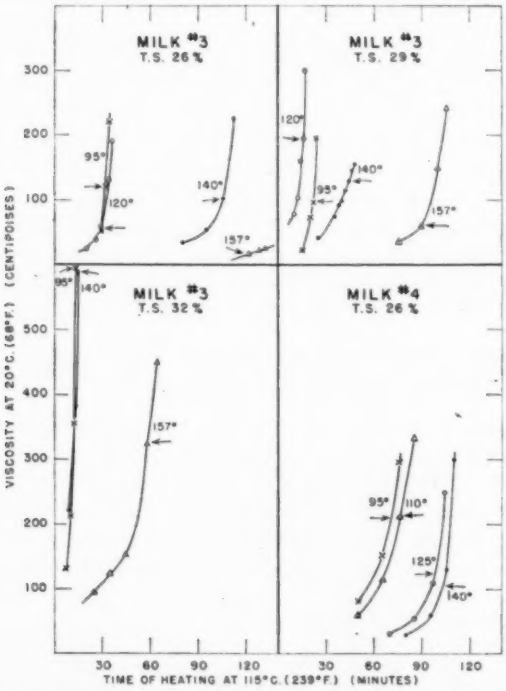

FIG. 2. The relationship between forewarming temperature, heat stability and the development of viscosity in two evaporated milks during sterilization. The solids content of the milk is indicated in each quadrant. The forewarming temperature is given for each curve; the forewarming time of the 95° C. samples was 10 minutes while for the other samples it was 25 seconds. The arrows indicate the time of the appearance of visible coagulation.

were: a thinning of all samples during early storage, then a period of only slight viscosity change, and finally a thickening in some of the milks.

The initial loss in body in all the milks tested was retarded by low storage temperatures. Samples of a representative 26 per cent solids milk held 55 days at 25° C. decreased in viscosity to the same extent as other samples held 8 days at 40° C. Samples held 55 days at 16° C. had the same viscosity as those held 5 days at 25° C.

Data on the effect of aging whole milk upon the viscosity of its 26 per cent evaporated milk were obtained. Fresh milk was held at 37° C. for 2 hours during which time the acidity increased from 0.18 to 0.21 per cent. Aging the fresh milk affected the viscosity of its evaporated product indirectly through its effect upon heat stability. Thinning of the milks during storage did not appear to be caused by the aging treatment.

The differences in the kind of body which may be expected in storage samples of evaporated milk of approximately 30 per cent solids content is

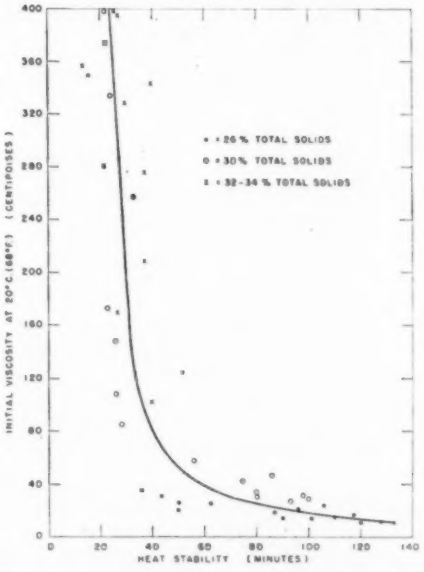

FIG. 3. Viscosities of evaporated milks of different heat stabilities measured after completion of the normal sterilizing process. Each milk was cooked for 18 minutes at 115° C. (239° F.), cooled to 20° C. and the viscosity determined within 3 hours after processing.

shown in figure 4. Milk A showed progressive thickening after completion of the initial thinning reaction, while milk B remained substantially unchanged in viscosity during the storage period.

Data on forewarming treatment, solids content, heat stability and viscosity of six batches of milk are given in table 1. The samples were stored and viscosity measurements were made at 30° C. The loss in the viscosity of these samples after 10 days' storage at 30° C. averaged 41 per cent of the original viscosity values. There was an additional average loss of 10 per cent of the original values during storage between the 10-day and 30-day periods. These losses are of the same order as those reported by Mojonnier and Troy (4).

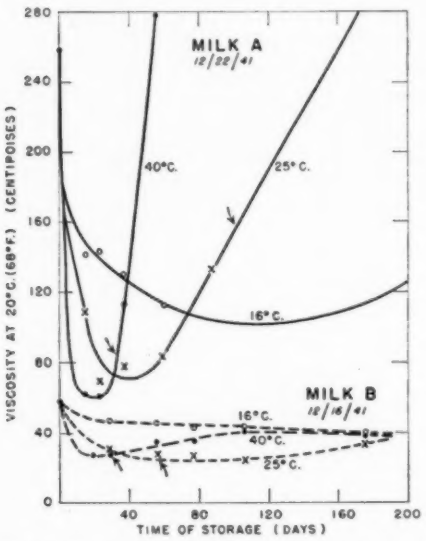

FIG. 4. The effect of time and temperature of storage upon the viscosity of two evaporated milks. The time of observance of heavy fat separation is indicated by arrows. Only moderate fat separation was noted in the 16° C. samples at the end of the storage period. Sample 12/16/41 was forewarmed at 140° C. (284° F.) for 30 seconds, contained 30.0 per cent solids and had a heat stability of 56 minutes. Sample 12/22/41 was forewarmed at 130° C. (266° F.) for 30 seconds, contained 31.7 per cent solids and had a heat stability of 33 minutes.

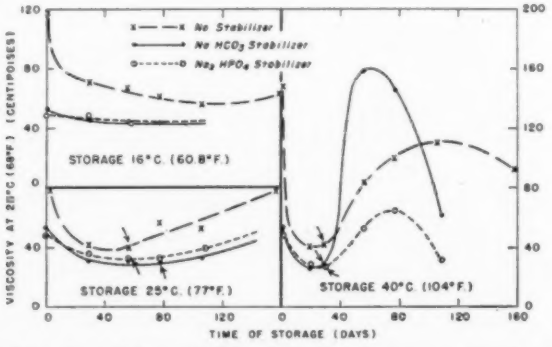

FIG. 5. The effect of time and temperature of storage upon the viscosity of samples of an evaporated milk sterilized with and without stabilizers. The time of observance of heavy fat separation is indicated by arrows. Only moderate fat separation was noted in the 16° C. samples at the end of the storage period. The milk was forewarmed at 95° C. for 10 minutes before concentration to 30 per cent solids content. The heat stabilities of the samples were: no stabilizer—26 min., bicarbonate stabilizer (4½ oz. per 1000 lbs. milk)—34 min., phosphate stabilizer (18 oz. per 1000 lbs. milk)—37 min.

The effect of large quantities of two stabilizing salts on the body changes of an evaporated milk during storage at three temperatures is shown in figure 5. This milk was representative of six samples upon which data of this nature were obtained. The high viscosity of the control sample can be attributed to its low stability.

Observations on fat-separation during storage were made on the evaporated milk samples in conjunction with the viscosity determinations. Figure 6 shows in a general way the relationships between temperature and time of storage. The curve for excessive fat-separation was based on visual observations; exact measurements were not made. If it is considered that cans of evaporated milk should be turned when they have stood half the time

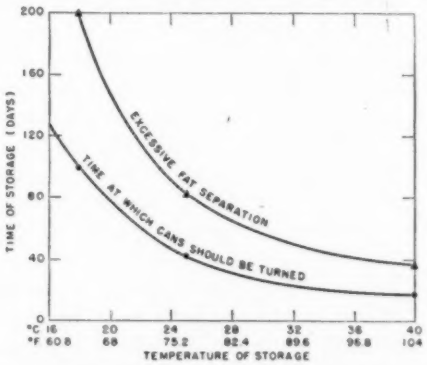

FIG. 6. The effect of temperature and time of storage upon fat separation in evaporated milks. The upper curve is based on average values from 20 milks of 26–32 per cent solids content held in undisturbed storage at different temperatures. The lower curve is an estimate of the time at which cases of evaporated milk in storage should be turned.

necessary to cause excessive fat-separation, the lower curve of figure 6 may be drawn. It would, therefore, seem reasonable to require the turning of cases of evaporated milk at some regular interval which lies below the lower curve of figure 6. For example, at 25° C. (77° F.) the milk should be turned every 6 weeks.

DISCUSSION

The viscosity of evaporated milk passes through well-defined changes arising from the manufacturing processes and the storage conditions to which it is subjected. There is first a thickening during the heat of sterilization, then a thinning during early storage and finally, after long periods of storage, the viscosity may again increase until a gel has formed.

The rate of thickening of evaporated milk during the sterilization process is variable, being the greatest shortly after heating is begun and immediately before coagulation appears. While the rapid thickening immediately pre-

ceding visible coagulation is, no doubt, a part of the coagulation process, all milks do not attain the same body before coagulation begins. The attainable body at coagulation seems to depend primarily upon the heat stability of the milk. Heat stability, in turn, is largely influenced by the concentration of milk solids, the conditions of forewarming, and the ionic equilibrium which exists in the serum.

The results of a number of experiments with milks differing in quality and forewarmed at different temperatures indicate that the relationship between the attainable viscosity at coagulation and the heat stability of the evaporated product is of a reciprocal nature.

Thickening in evaporated milks during sterilization does not proceed rapidly until about 10 minutes before coagulation. A processing period of about 20 minutes is generally favored in commercial practice. If a heavy, creamy body is to be developed, the heat stability of the milk should not exceed 30 to 40 minutes. When the cooking time to be used approaches the heat-stability time, extensive increases in viscosity may be encountered. Figures 1 and 2 show the thickening for which each minute of heating may be responsible when the milk is in the precoagulation phase. Milks with stabilities in excess of 50 minutes will be exceedingly thin after heating them at 115° C. for 20 minutes unless an increase in solids content is depended upon to build body.

The first change which occurs in the body of evaporated milk during storage is a rapid thinning, an effect which is accelerated at high storage temperatures. As thinning progresses a viscosity level is finally reached which may be termed the basic storage viscosity. In this region the viscosity ceases to decrease so rapidly; there is a distinct flattening in the viscosity curve (figs. 4 and 5, and table 1). As the milk loses body, another storage change, a separation of the fat, begins.

During storage, control of the body of evaporated milk should be first concerned with methods for delaying the initial thinning process and establishing a relatively high basic storage viscosity. Attainment of the basic storage viscosity may be effectively delayed by subjecting the milk to low storage temperatures. When evaporated milks were stored 60 days at 20° C. (68° F.) or lower, the loss in viscosity was less than the loss which occurred after 10 days at 40° C. (104° F.). Evaporated milks subjected to storage temperatures above 30° C. (86° F.) often reached their basic storage viscosity in 10 days.

Although the level of the basic storage viscosity was determined largely by heat stability, it was also influenced by the solids content of the milk. The 37 per cent solids milk of June 25, table 1, had a stability of 35 minutes and a viscosity after 30 days' storage of 121 centipoises. A portion of the same milk forewarmed at 120° C. (with no holding time) to give the same stability (34 minutes) at 27 per cent solids content had a viscosity of only 15 centipoises after 30 days' storage.

No manufacturing treatment, such as alteration in the type or conditions of forewarming, has yet been found which proved capable of preventing the initial rapid loss in the viscosity of evaporated milk during storage at high temperatures.

The last stage in body deterioration of evaporated milk consists of a thickening of the milk, which sometimes reaches the point of gelation. Storage thickening is rarely found in the commercial product, partly because much of the milk is consumed before thickening sets in and partly because some milks apparently never would reach this phase of development. Before thickening becomes objectionable an excessive fat-separation can generally be observed. The viscosity increase during storage usually starts too late to be very helpful in retarding fat-separation.

Certain samples, such as milk B, figure 4, did not thicken. The storage viscosity curves of many 26 per cent evaporated milks followed the course of milk B. High-solids milks thickened more readily than did 26 per cent milks but the greatest increase occurred in the solids range above 32 per cent.

The use of sodium bicarbonate as a heat stabilizer (fig. 5) accelerated storage thickening in several milks but not before a heavy layer of fat separated from the samples. Sodium bicarbonate is no longer used as a stabilizer in the manufacture of evaporated milk.

Evaporated milks which have been subjected to sterilization processes requiring only a minute or two to complete and consequently which have little cooked flavor and color are known to thicken quickly during storage. Evidently the drastic heat treatment of the commercial product retards storage thickening while the mild treatment given various experimental batches permits it.

Little is known of the real cause of thickening in evaporated milk during storage. It may be a slow continuation of the coagulation process accompanied by an orientation of the caseinate molecules which finally produces an irreversible gel structure. Molecular orientation is suspected as being involved because undisturbed storage appears to promote thickening and the early incipient gel structure may be easily reduced by shaking.

It is hoped that future investigations on the viscosity of evaporated milk will eventually indicate a means of controlling the thinning and thickening processes so that a good body in evaporated milk can be produced and permanently held during storage.

SUMMARY

1. The viscosities of evaporated milks of 26 to 36 per cent solids concentrations followed a well-defined pattern during processing and storage. Thickening occurred during the sterilization process; this was followed by a loss of body and a thinning early in the storage period. The low storage viscosity was maintained for various and unpredictable lengths of time, dur-

ing which fat-separation occurred. Late in the storage period a final thickening, which often proceeded to gelation, sometimes set in.

2. The rate of thickening of evaporated milk during sterilization at 115° C. (239° F.), although variable, becomes greatest in the 10-minute period immediately preceding coagulation. A heavy, creamy body could be developed during this pre-coagulation stage. In general, the lower the heat stability of a milk, the higher the viscosity it would develop during the 10-minute period preceding coagulation. Concentrated milks of high heat stability which reached the end of the sterilization period before entering the thickening phase did not develop, during processing, the high viscosity shown by milks of lower heat stability.

3. The body formed during the sterilization of milks of different concentrations became greater as the solids content of the milk was increased. The heaviest viscosities were produced in milks of high concentration and low heat stability.

4. Variations in milk quality and in forewarming procedures affected the viscosity of evaporated milk indirectly through their effect upon the heat stability of the milk.

5. The magnitude of the decrease in the viscosity of evaporated milk during early storage was dependent upon the storage temperature. At temperatures below 16° C. (60.8° F.) the loss of body was small. At temperatures above 30° C. (86° F.) evaporated milk sometimes lost 40 per cent of its original viscosity during the first 10 days of storage.

6. After the initial thinning in the viscosity of evaporated milk held in storage at constant temperature, a basic storage viscosity level was reached beyond which the rate of viscosity loss during prolonged storage was small.

7. Some evaporated milks, especially those receiving light heat treatments and those with a high concentration of solids, began to show increases in viscosity even to the point of gelation, late in the storage period.

8. The results indicate that the procedure for developing and maintaining a satisfactory body in evaporated milk which is used by many manufacturers conforms to the best practice that can be devised from our present state of knowledge. Assuming efficient homogenization and uniform handling of the canned product, important steps in body control are: (a) adjust the heat stability to a value of a few minutes greater but not more than twice the cooking time, bearing in mind that viscosity increases rapidly during the 10 minutes preceding coagulation; (b) store the finished milk at a temperature below 21° C. (70° F.); (c) turn the cases approximately every six weeks; (d) the viscosity may be increased by raising the milk solids content of the product.

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REFERENCES

(1) GRINDROD, GEORGE. Sterilization of Milk by Impact of Steam. Creamery and Milk Plant Monthly, 16: 9, 38. 1927.

(2) HUNZIKER, O. F. Condensed Milk and Milk Powder. Ed. 5, p. 194. 1935

(3) MALLORY, W. E. Ann Arbor, Michigan. U. S. Patent #2,270,540. 1942.

(4) MOJONNIER, T., AND TROY, H. C. The Technical Control of Dairy Products, Chapter 19, p. 781. 1925.

(5) WEBB, B. H., AND BELL, R. W. The Effect of High-Temperature Short-Time Forewarming of Milk upon the Heat Stability of Its Evaporated Product. JOUR. DAIRY SCI., 25, No. 4: 301. 1942.

(6) WEBB, B. H., BELL, R. W., DEYSHER, E. F., AND HOLM, G. E. The Effect of Various Degrees of Forewarming upon the Heat Stability of Milk of Different Solids Concentration. JOUR. DAIRY SCI., 26, No. 7: 571. 1943.

THE BACTERIOLOGY OF BULL SEMEN. II. THE EFFECT OF BACTERIA UPON RAPID TESTS FOR SEMEN QUALITY

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In a recent report Beck and Salisbury (1) have proposed two rapid methods for estimating the quality of bull semen; namely, the rate of methylene blue reduction by semen in yolk-citrate diluent and the survival of spermatozoan motility during short-time-high-temperature incubation. The following work was undertaken to determine the effect of bacterial contamination upon the accuracy and validity of these quick tests, especially the specific effect on the methylene blue reduction rate. The effect upon the methylene blue test was studied by two methods: first, by calculation of the multiple correlation coefficients to make allowance for the reduction due to spermatozoa and other known factors (1) in semen samples; and, second, by determining the methylene blue reduction time when pure cultures of bacteria were inoculated into yolk-citrate diluent, with and without seminal plasma. The effect of the short-time-high-temperature incubation test upon the bacterial content of semen has also been determined.

A preliminary estimate of the effect of bacteria upon the methylene blue reduction test may be obtained by an examination of the data available for other biological fluids, as for example milk. Thornton and Hastings (7) using 1:200,000 methylene blue at 37° C. found the bacterial count at the time of complete reduction to range from 3.5 to 45 million bacteria per ml. No sample with an initial count of fewer than 20 million bacteria per ml. gave a reduction time of less than one-half hour. In this and similar work the factors shown to affect the reduction time are the type of bacteria, their physiological condition, the temperature, the nature of the substrate present, and the concentration of methylene blue to be reduced. That storage may also be a factor was shown by Fayer (3) who stored milk samples at 4° C. for 18–72 hours and found a decrease in the rate of methylene blue reduction without a change in bacterial count. Bacterial counts of the magnitude usually found in semen samples would not on the basis of the findings of Thornton and Hastings (7) be expected to affect the proposed methylene blue reduction test.

EXPERIMENTAL

Methods

The semen samples used in these experiments were obtained under routine conditions either from bulls in the dairy herd at Cornell University

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or from bulls owned by the New York Artificial Breeders' Cooperative in Syracuse, New York. All semen samples were collected with the artificial vagina. The semen was handled during examination, dilution and preparation for storage after the methods described by Willett and Salisbury (8). The methylene blue reduction tests were carried out as previously described (1). Briefly the test involves an observation of the time required for the complete reduction of 1:40,000 methylene blue by bull semen diluted at a constant rate with yolk-citrate diluent. With the exception of the experiments to determine the effect of various temperatures on the reduction time and upon bacterial numbers, the tests were run at 45° C. Bacterial counts were made on 2 per cent blood agar plates incubated 3 days at 37° C.

The effect of bacteria present in semen on the reduction test

For determination of the effect of bacteria, freshly diluted semen samples were selected from the bulls found in earlier studies (4) to yield semen containing the highest bacterial counts. Semen from these bulls was also of poorer than average quality in other respects; *i.e.*, long methylene blue reduction times had been noted. These samples were selected as those most likely to give erroneous estimates of semen quality if bacteria have a significant effect upon the methylene blue reduction test.

A factorial experiment was designed to simultaneously study the effect of the number of bacteria on the methylene blue reduction time at several temperatures and the effect of a 45-minute incubation period at these temperatures on the bacterial count. Samples of the same ejaculates were stored for 2, 4, 6, 8, and 10 days, following which bacterial counts and methylene blue reduction tests were run. Aliquots of the same samples were incubated and stored with and without methylene blue. In this way the effect of high-temperature incubation, apart from methylene blue, upon the bacterial count was determined. The methylene blue was added to one series of stored samples with the thought that if the reduction time was not influenced by bacterial growth, or other factors, the relative metabolic activity of the semen could be determined at any time during storage by simply warming a sample to the proper temperature and observing the reduction time.

For each ejaculate the spermatozoan concentration was determined at the time of collection and the motility was observed immediately before each methylene blue reduction test was performed. Thus, it was possible to determine the multiple effects of storage interval, spermatozoa count, motility, and bacterial count on the time required to reduce methylene blue. Ten ejaculates were used and 21 individual plate counts, motility estimates and methylene blue reduction tests were performed on each diluted ejaculate. It was later found necessary in the statistical analysis of the storage experiment to remove the data for two ejaculates as the semen was of such poor quality that it did not reduce the methylene blue in the time allotted for the

incubation period. The removal of these data had no influence on the conclusions drawn from these experiments.

For fresh samples the correlation coefficients between methylene reduction time for each of the following was: spermatozoan concentration, – 0.7780; spermatozoan motility, 0.3431; and bacterial count, – 0.1427. The coefficient for spermatozoan concentration is the only significant observation (n = 10). Though these fresh samples contained a wide range of bacterial counts the number present have but a minor influence on the methylene blue reduction time. The stored samples showed even wider variation in bacterial count. With these samples the calculated correlation coefficients for each of the following was: spermatozoan concentration, – 0.6126; spermatozoan motility, – 0.3110; and bacterial count, – 0.1022 (n = 48). The first two coefficients are statistically significant, the last is not. Even with the larger number of observations the data indicate that the part played by bacteria in the reduction time is not large. All samples contained predominantly diphtheroid organisms, except one ejaculate in which the majority of the organisms were *Pseudomonas aeruginosa* (*pyocyaneus*).

The mean data for the fresh and stored samples are shown in tables 1 and 2. The methylene blue reduction times for fresh semen are too short for bacterial multiplication to be an important factor, when the tests are run at temperatures above 45° C. as recommended. In fact, the experiments show (table 1) a decrease in bacterial count at temperatures of 45° C. and above. The decrease is especially marked in the presence of methylene blue.

The stored samples (table 2) showed, on the average, a slight decrease in bacterial count over the first 4 days followed by an increase. Over this period, the methylene blue reduction time increased slightly and then decreased. The correlated changes are, however, too slight to be of significance.

The effect of methylene blue on the numbers of bacteria

From table 1 a comparison of the numbers of bacteria found in semen samples after 45 minutes incubation with and without methylene blue may be obtained. The samples used had stood at the laboratory temperature for about 30 minutes before incubation, which time was found to be the average required to set up the experiments. In table 2 may be seen a similar comparison of bacterial counts made on the same ejaculates after storage at 5° C. The methylene blue had been added to one series of samples before storage, and to the other just prior to the 45-minute incubation period. As limited reduction had proceeded in the samples to which methylene blue was added before storage, the reduction times were shorter than for the samples to which the reagent was added just before incubation.

It will be noted that incubation of the fresh semen in the presence of methylene blue reduced the bacterial count in all cases. In the absence of methylene blue, the count decreased only at the two higher temperatures. An analysis of variance of the data showed these differences to be highly

TABLE 1

The effect of incubation temperatures on the bacterial count of semen samples

No. samples	Spermatozoa	Motility fresh	Bacterial count	Mean methylene blue reduction time during and mean bacterial counts after 45 minutes' incubation at									
				37.5°		40.0°		42.5°		45.0°		47.5°	
				Time	Count	Time	Count	Time	Count	Time	Count	Time	Count
	thousands per mm.^{3}	*%*	*thousands per ml.*	*min.*	*thousands per ml.*	*min.*	*thousands per ml.*	*min.*	*thousands per ml.*	*min.*	*thousands per ml.*	*min.*	*thousands per ml.*
8	1109	66.2	640	31.5	553	29.6	624	12.1	561	9.7	463	9.9	316
				Mean bacterial counts after 45 minutes' incubation without methylene blue present									
				.....	879	.....	897	.....	884	.....	590	.....	503

TABLE 2

Mean reduction time and bacterial count after storage at 5° C.

Storage period	Spermatozoa motility	Mehylene blue reduction time	Bacterial count after 45 minutes at 45° C.	
			Without methylene blue	Methylene blue added before storage
days	*%*	*min.*	*thousands per ml.*	*thousands per ml.*
0	66.2	9.7	640	640
2	47.5	14.7	420	363
4	37.5	12.9	307	237
6	31.2	11.1	1,141	567
8	30.0	12.1	3,171	1,054
10	25.0	13.2	2,888	2,279

significant. These facts support the use of 46.5° C., previously suggested by Beck and Salisbury (1), for the short-time-high-temperature incubation and methylene blue reduction tests. These workers have shown a very high correlation between the decrease in motility at 46.5° C. in 1 hour and the decrease in motility during 10 days' storage at 5° C. The bacterial count for samples stored with methylene blue was slightly lower in all cases than samples stored without methylene blue, but the difference was so small as to suggest that this was only a chance deviation. In the previous report (1) it was shown that the amount of methylene blue used did not have a detri-

TABLE 3

Methylene blue reduction test on pure cultures of bacteria in yolk-citrate diluent

Organism added	Days stored at 5° C.					
	0		5		10	
	Bacterial count	Reduction time	Bacterial count	Reduction time	Bacterial count	Reduction time
	millions per ml.	*min.*	*millions per ml.*	*min.*	*millions per ml.*	*min.*
Escherichia coli	130.0	.....	0.6	.....	11.0	100.0
	1,500.0	31	15.0	40	13.0	> 120.0
Aerobacter aerogenes	900.0	> 120	48.0	.....	320.0	7.5
			750.0	11	1,000.0	4.5
Pseudomonas aeruginosa	800.0	> 120	750.0	.....	36.0	10.0
			9.0		120.0	6.0
Staphylococcus aereus	1,200.0	> 120	0.18	.....	9.0	14.0
			15.0	.....	6.0	.....
Diphtheroid strain 1	2.0	.....	0.13	.....	0.3	.....
2	7.0	.....	21.0	.....	4.0	.....
	thousands per ml.	*min.*	*thousands per ml.*	*min.*	*thousands per ml.*	*min.*
None (control)	0.03	.....	0.3	.....	4.0	.....

mental effect on the maintenance of spermatozoan motility in stored samples. From these observations it has been suggested that this dye could be used to identify the semen of one breed of bulls from that of another and thus avoid field errors which sometimes arise in artificial insemination.

The effect of specific types of bacteria on methylene blue reduction in yolk-citrate diluent

To test directly the effect of bacteria, yolk-citrate diluent and yolk-citrate diluent plus seminal plasma were inoculated with pure cultures in numbers higher than those usually found in semen and the methylene blue reduction test performed at 45° C. These tests were, as previously mentioned, performed within 30 minutes of inoculation. Other inoculated samples were stored for 5 and 10 days before reduction tests were performed. Twenty-

hour cultures of organisms isolated from semen samples were used as inoculum. These organisms were encountered in large numbers in various samples of semen during the course of the study. The data in tables 3 and 4 indicate that in fresh samples, none of the organisms would interfere with the test even when present in numbers as high as 1 billion per ml. The shortest reduction time (30 minutes) which was obtained with 1.5 billion *E. coli* was still beyond the reduction time for all but the poorest semen samples.

The results with samples stored for several days were not as satisfactory. If bacteria, which grow at the storage temperature, are present the reduction

TABLE 4

Methylene blue reduction test on pure cultures of bacteria in yolk-citrate plus seminal plasma

Organism added	Days stored at 5° C.					
	0		5		10	
	Bacterial count	Reduction time	Bacterial count	Reduction time	Bacterial count	Reduction time
	millions per ml.	*min.*	*millions per ml.*	*min.*	*millions per ml.*	*min.*
Escherichia coli	48.0	60	54.0	48	75.0	9.0
	6.0	.....	7.0	.....	.....	.....
Aerobacter aerogenes	17.0	120	900.0	12	2,000.0	6.0
Pseudomonas aeruginosa	60.0	120	66.0	120	60.0	6.5
Staphylococcus aureus	8.0	120	30.0	120	20.0	16.0
Diphtheroid strain 1	3.0	.....	6.0	.....	4.0	15.0
2	0.02	.....	0.03	.....	0.05	40.0
	thousands per ml.	*min.*	*thousands per ml.*	*min.*	*thousands per ml.*	*min.*
None (control)	0.015	.....	0.009	.....	.....	.....

time may be very short. Cultures of *Aerobacter aerogenes* and *Pseudomonas aeruginosa* not only grow at 5° C. but the citrate present in the diluent is an excellent substrate for these organisms. When 750 million or more *Aerobacter aerogenes* organisms per ml. are present (tables 3 and 4), they would interfere markedly with the validity of the methylene reduction test after 5 days' storage at 5° C. After 10 days' storage counts of a hundred million of either of these two species of bacteria reduced the methylene blue fast enough to interfere with the test. When the seminal plasma was added to the yolk-citrate short reduction times were observed with each organism after a storage period of 10 days. The effects of the bacterial types contributed by the seminal plasma were not determined. However, as one may observe from the tables the number of bacteria in the plasma was very low. The range of bacterial numbers, in fresh diluted semen, the mean of which is shown in table 1, was from 97 thousand to 4.5 million.

After 10 days' storage the average count had increased to about 3 million bacteria per ml. with one count as high as 60 million. In only three diluted semen samples were the numbers as high after 10 days' storage as was found in the inoculated material. In three ejaculates the count increased as much as 20-fold, while in three others the total count decreased during storage. Greater increases in count were reported in a recent publication of the United States Department of Agriculture (5). With higher initial counts these workers report a 10-fold increase in bacterial count during 7 days and a 65-fold increase during 10 days' storage at 4° C.

A study of the data of this experiment reveals that only part of the short reduction times observed for stored yolk-citrate may be attributed to bacterial numbers. For diluent, or diluent plus seminal plasma, stored 10 days, short reduction times often were found where the numbers of bacteria present was no higher than those which in fresh yolk-citrate required more than 2 hours to reduce the dye. Apparently some factor other than numbers of bacteria present is partially responsible for the short reduction times observed for stored, inoculated material.

In this connection it should be pointed out that the negative correlation between spermatozoa count and methylene blue reduction time continued at the high level previously reported (1) regardless of the length of storage interval. Such definite trends were not noted for the relation between motility of the spermatozoa and methylene blue reduction time as the storage interval advanced. For the present, the methylene blue test for quality of semen should be used only with fresh semen and fresh diluent for the most satisfactory results.

DISCUSSION

From the results of these investigations it is concluded that if bull semen is collected and handled with proper sanitary precautions, and if the diluent is prepared following a definite aseptic routine (4) the number of bacteria in fresh semen and diluent will not interfere with the estimates of semen quality obtained with the methylene blue reduction test. In addition, the temperature recommended for conducting the test, 46.5° C., is high enough that bacteria are unable to grow. In fact, such a temperature has a lethal effect on the bacteria, especially in the presence of methylene blue.

Of the bacteria most likely to interfere with the test, the coli-aerogenes organisms can be largely excluded by sanitary care in preparation of equipment and during collection and handling of samples. The exclusion or control of the number of pseudomonas organisms is more difficult. These bacteria are sometimes found in large numbers and as the predominating type in the semen of some bulls. Ordinary methods of control do not influence the numbers of these bacteria for their source appears to be deep in the reproductive tract. Bulls harboring *Pseudomonas aeruginosa* in their reproductive tracts are likely to have poor breeding records (2, 4) and are poor risks for artificial insemination.

For stored samples of diluted semen the methylene blue reduction test should be used with caution. Certain unknown factors, in addition to the apparently minor effects of the number and kinds of bacteria, influence the test with semen samples stored more than 2 days so that reliable conclusions may not be drawn. For estimation of the quality of samples stored for 4 or more days observation of the duration of motility at 46.5° C. should give reliable estimates of potential livability of spermatozoa during continued storage at 5° C. Further work needs to be done before the factors influencing the reduction test in samples of diluted semen stored more than 2 to 4 days can be established.

SUMMARY

1. When sanitary precautions are observed, the number of bacteria found in freshly drawn semen or freshly prepared yolk-citrate diluent is not sufficient to interfere with the methylene blue reduction test for semen quality.

2. The short-time-high-temperature incubation test for semen quality may kill up to 50 per cent of the bacteria present when the test is run at 45° C. or above in the presence of methylene blue. A temperature of 46.5° C. is recommended.

3. The methylene blue reduction test is not recommended as a criterion of the quality of semen stored more than 2 days.

4. The short-time-high-temperature incubation test is recommended as a criterion of continued livability of the spermatozoa in stored semen samples.

5. Though the bacterial population of stored samples can be controlled by proper precautions, factors other than the number of bacteria, and the activity and concentration of the spermatozoa are involved in the methylene blue reduction rate of stored samples.

REFERENCES

(1) Beck, G. H., and Salisbury, G. W. Rapid Methods for Estimating the Quality of Bull Semen. Jour. Dairy Sci., 26: 483–494. 1943.

(2) Campbell, J. J. R., Gunsalus, I. C., and Salisbury, G. W. Unpublished Data. 1943.

(3) Fayer, J. M. Light and Temperature as Factors in the Methylene Blue Reduction Test. Vt. Agr. Expt. Sta. Bul. 374. 1934.

(4) Gunsalus, I. C., Salisbury, G. W., and Willett, E. L. The Bacteriology of Bull Semen. Jour. Dairy Sci., 24: 911–919. 1941.

(5) Report of the Chief of the Bureau of Dairy Industry. U. S. Department of Agriculture, p. 16. 1942.

(6) Salisbury, G. W., Fuller, H. K., and Willett, E. L. 1940. Preservation of Bovine Spermatozoa in Yolk-citrate Diluent and Field Results from Its Use. Jour. Dairy Sci., 24: 905–910. 1941.

(7) Thornton, H. R., and Hastings, E. G. Studies on Oxidation-Reduction in Milk. The Methylene Blue Reduction Test. Jour Dairy Sci., 13: 221–245. 1930.

(8) Willett, W. L., and Salisbury, G. W. The Effect of Various Diluters, Cooling Rate, Temperature of Storage, and Some Other Factors on the Livability of Spermatozoa in Stored Samples of Bull Semen. Cornell Univ. Agr. Expt. Sta. Memoir 249. 1942.

PEPPERGRASS SILAGE

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Recently attention has been directed toward the use of weed crops for forage purposes. At the Virginia Station alfalfa and brome grass were established to serve as supplementary pasture crops to be available in July and August when bluegrass is in low production. The alfalfa had been pastured off by May 20th and there was a heavy stand of peppergrass (*Lepidium virginicum*) remaining unpastured. There was entirely too much of this weed to permit it to go to seed. If clipped and allowed to remain on the ground the peppergrass would smother out the alfalfa. Mowing, raking and hauling away the peppergrass would be costly if no use were made of it.

Ten acres of peppergrass, yielding about 28 tons, were cut and ensiled with 50 pounds of molasses per ton as a preservative after due consideration of the possibility that the silage might be strong with peppergrass flavor.[1] Five acres were cut from the alfalfa field and some alfalfa was picked up by the mowing machine. The other five acres were cut from the brome grass field and some brome grass was clipped. It was estimated that the ensiled material was 80 per cent peppergrass. The seeds were immature but had the field been mowed and the peppergrass left on the ground the seeds would probably have matured sufficiently to germinate. When the material was cut into the silo there was sufficient volatile material in the air to irritate the eyes. The ensiled material kept well. After the silage had undergone fermentation there was no trace of the peppergrass flavor or odor remaining. Cows ate up to 50 pounds per day of the silage and appeared to relish it better than silage made from bluegrass with corn meal as a preservative. Samples of peppergrass in as nearly as possible the same stage of maturity as that which was ensiled were submitted to the Agricultural Chemistry Department for analysis. A sample of peppergrass silage containing about 20 per cent alfalfa was taken from the silo and submitted also. The analyses of these materials are shown in table 1 with the analysis of corn silage for comparison. Table 2 shows that the quality of peppergrass silage was satisfactory as there was no decline in production due to peppergrass silage. Cow number 6 was nearly dry and therefore any change in the feed would not have prevented a decline in the milk production of the cow.

The milk had no taste of peppergrass or indole. When the milk was

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[1] Hussong, R. V., and Quam, Sidney. Relationship of Consumption of Peppergrass by Cows to the Flavor and Indol Content of Butter. Jour. Dairy Sci., 26, No. 6: 505–513. 1943.

TABLE 1

Analyses of peppergrass and peppergrass silage compared with corn silage

	Corn silage* (dent, well matured, well eared)	Peppergrass	Peppergrass silage
	%	%	%
Dry basis			
Protein	8.56	12.86	16.19
Fat	3.08	5.54	4.40
Crude fiber	21.92	34.17	23.63
Ash	5.48	9.32	.....
Nitrogen-free extract	60.95	38.11	40.62
Total dry matter	29.20	36.47	28.75
Phosphorus	0.21	0.23	0.42
Calcium	2.74	1.34	1.21
Potassium	0.96	3.00	2.41
Magnesium	.....	0.40	.....
Silica	.....	1.28	.....

The peppergrass was analyzed by Mr. H. H. Hill and the peppergrass silage by James F. Eheart, both of the Agricultural Chemistry Department. The peppergrass contained approximately 20 per cent alfalfa.

* The values on the dry basis are computed from data of Morrison's Feeds and Feeding, 20th Edition.

TABLE 2

The milk production of cows fed bluegrass silage and peppergrass silage

Cow No.	Bluegrass silage		Peppergrass silage	
	Amounts of milk produced			
	May 2–16	May 17–31	June 1–15	June 16–30
	pounds	*pounds*	*pounds*	*pounds*
1	393.0	350.0	313.5	332.5
5	476.5	429.0	431.5	455.0
6	274.0	183.0	145.5	108.0
7	400.0	346.5	321.0	336.5
Average daily milk flow for 4 cows	27.6	23.4	21.6	22.0

The rate of decline of milk flow was reduced when bluegrass silage was replaced with peppergrass silage.

TABLE 3

The quality of butter made from milk produced by cows fed peppergrass silage

Lot No.	Age of cream in days	Condition of cream at churning	Butter
1	7	High acidity	Salted, old cream flavor, no peppergrass flavor
2	7	Low acidity	Unsalted, superior flavor, no peppergrass flavor
3	1	Sweet	Unsalted, excellent flavor, 92 score

separated the cream had a normal flavor. Butter was made from the cream without using a starter with the results shown in table 3.

Three judges scored the butter samples and no off-flavors attributable to the peppergrass were detected.

Evidently the substances producing the peppery flavor of the green material were oxidized in the fermentation process. The growing of peppergrass for silage is not advocated; however, crops that contain peppergrass may be ensiled without fear that dairy products will have an off-flavor as a result of their use.

One hundred peppergrass seeds were taken from the silage two weeks after ensiling for a germination test. There was no germination. Furthermore there was no germination of seed taken from the silo in early September, after the elapse of a normal rest period for these seeds.

SUMMARY

Ensiling peppergrass (*Lepidium virginicum*) with molasses as a preservative produced a palatable silage which after fermentation had no peppergrass flavor. Neither cream nor butter produced from the milk of cows fed this silage showed any off-flavor. Peppergrass seeds did not germinate two weeks after ensiling. Crops containing peppergrass may be ensiled without fear that off-flavor will result in the milk or milk products.

ACKNOWLEDGMENT

The assistance of Dr. C. C. Flora in producing the butter and judging the butter samples is gratefully acknowledged.

A NEW QUANTITATIVE METHOD FOR ESTIMATION OF TOTAL COMBINED LENGTH OF MOLD FRAGMENTS IN BUTTER[1]

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The method described in this report resulted from studies on retention of mold fragments (mycelia)[2] by butter, buttermilk and wash water during manufacture of butter.

During these and previous studies (3) it became apparent that a more quantitative method for estimating content of mold hyphae or filaments in butter and other products would have to be developed before information could be obtained on mold retention by the various materials studied. Since the "mold mycelia value" of butter by the official method now in use is influenced primarily by the total mass of mold growth present, the logical quantitative method would be one that would measure the total length of all combined mold fragments in a measured sample. The difficulty of extracting the mold fragments renders a macroscopic approach impossible. The possibility of analyzing for some chemical constituent contained by the mold hyphae and foreign to butter appears remote at the present time. About the only possibility remaining is a microscopic method. The Wildman method (4) in use at present provides a comparative rather than quantitative value because it does not measure the total length of the combined filaments in a sample. A method was therefore developed in which the total combined length of the mold hyphae visible with low power (100×) magnification of the microscope was determined and expressed in millimeters per gram or mgm. Because the method has proven so convenient for certain studies on mold content of butter and other materials, it is being published with the hope that it may prove useful to other investigators in this field.

DESCRIPTION OF METHOD

1. Thoroughly mix 1 g. butter with 9 ml. hot (70–75° C.) gum solution. (It is desirable to scrape off about one-eighth inch of the surface butter and

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[2] The term "mold mycelia" now commonly used in referring to the amount of dead mold filament in butter should perhaps, technically speaking, be changed to "mold hyphae" or "fragments of mold hyphae." A mold hypha (plural—hyphae) is a single thread or filament of mold. A mold mycelium (plural—mycelia) consists of a mass or network of interlacing filaments or hyphae. Therefore it is the filaments or hyphae or hyphae fragments that are counted in the Wildman method. Filaments, hyphae and mold fragments are used synonymously in this paper.

then obtain the one gram sample by scraping it from an area of a few square inches.)

2. Transfer 0.1 ml. to one side of a 1×3 inch (26×77 mm.) slide.

3. Spread over exactly one-half of the slide. (One means of evenly distributing the preparation is to spread the material with a small, solid glass rod, first lengthwise and then crosswise. The preparation seems to distribute itself more evenly if, after spreading on the slide, it is placed for a few minutes on a level surface at room temperature before setting on a warm surface to dry.)

4. Dry on a warm surface.

5. Stain with Newman's (1) or a Modified Newman's stain. (The same precautions regarding drying of stains before washing to remove excess dye, should be observed as in the staining of milk films with Newman's stain. It is possible to wash the slides too long in removing the stain. A few trials will indicate the amount required. Nevertheless some mold hyphae fragments will not stain intensely and a knowledge of the microscopic appearance of mold hyphae is helpful in identifying them.)

6. Calibrate factor for low power objective of microscope. Include dilution of sample and area of slide.

7. Examine 25 or preferably 50 fields on each slide. If the sample contains much mold, 25 fields may be sufficient. Estimate, by means of a ruled ocular micrometer disc, the total length of mold filament for each field. (Appearance of the preparation under the microscope is often improved by covering it with a thin film of immersion oil. This is particularly true where the method has been applied to substances containing considerable protein, such as cream or buttermilk.)

8. Calculate average length of filament per field.

9. Multiply average length of filament per field by the factor.

10. Express results as millimeters per gram or preferably millimeters per mgm.

SAMPLE CALCULATION

A. Diameter of field = 1.66 mm.

B. Area of field = πr^2 = 2.164 sq. mm.

C. Area of one-half slide = 1001 sq. mm.
Number of fields in one-half slide = 463.

D. Butter diluted 1 : 10; 0.1 ml. placed on slide.
Total dilution = 100 times.

E. Factor = C × D = 46,300.

F. Average total mm. mold filament per field of sample preparation = 0.346.

G. Mm. mold per gm. = E × F = 16,020.

H. Mm. mold per mgm. = $\frac{E \times F}{1000}$ = 16.02.

Some comments might be made on certain steps in the procedure. The diluting medium was a 0.75 per cent carob bean gum solution similar to that used in the Wildman method (4). Other gum solutions that provide sufficient viscosity and a reasonably clear solution should be satisfactory. Among the preparations also used in these studies were the common adhesives or fixatives used to fix tissue sections to slides. No difficulty was experienced with cracking, peeling or slipping-off of films when carob bean gum solution was used and the above staining procedure followed and slides rinsed gently in a container of water.

Numerous trials indicated that 1 g. of butter in 9 ml. gum solution and 0.1 ml. of this on a slide provided about the optimum dilution. A greater amount of butter increased the difficulty of fat removal and too small a sample provided too little mold for counting.

A number of common staining techniques were tried, including the Gram stain, negative staining, and staining with methylene blue after defatting in xylol. None of these showed any advantage over the simple Newman method used for milk smears. Substitution of half of the methylene blue in the Newman stain with basic fuchsin resulted in a preparation that stained the mold more intensely on some of the slides. A better staining technique for this purpose can possibly be developed.

For a sample of high mold content 25 fields per slide should be sufficient to count; however, 50 fields is preferable for a sample with moderate or low mold content. On the basis of present official standards for butter, 20 mm. per mgm. would be considered a high count. Duplicate slides should be prepared and results averaged.

RESULTS

A number of trials were carried out to determine the reliability of the new method. These included checks between various slides from the same sample, different samples from the same print of butter and counts on the same prints by two technicians using different microscopes with separate preparation of samples and slides.

TABLE 1

Results on different samples from the same prints of butter

Mm. mold per mgm. butter					
Sample A			Sample B		
Slide 1	Slide 2	Ave.	Slide 1	Slide 2	Ave.
12.87	11.20	12.04	10.19	11.85	11.02
3.80	3.43	3.62	3.80	3.52	3.66
8.15	9.72	8.94	12.13	8.89	10.51
2.87	3.89	3.38	3.33	2.96	3.15
20.46	19.54	20.00	19.08	18.06	18.57
25.74	23.15	24.45	25.56	25.09	25.33

TABLE 2

Counts with new method by two operators using different microscopes

Sample No.	Total combined length of mold fragments	
	A	B
	mm. per mgm.	*mm. per mgm.*
320	16.56	16.56
321	48.85	48.76
322	21.25	25.76
323	7.82	8.10
324	16.74	20.24
325	14.17	12.70
326	21.16	22.45
327	11.87	10.76

The values shown in table 1 are from a trial to determine the consistency of results between duplicate samples. As the results indicate, with one exception, the variation between the four slides on the two samples is relatively small for a microbiological analysis. The mold content of different portions of a print of butter appeared fairly constant. Whether different portions of a commercial churning would check as well with this method has not been determined. The results of Claydon (2) indicate that some significant variations may occur due to chance variation of mold filaments in different portions of a churning.

Two different operators employing different microscopes and separate preparation of samples can obtain comparable results with the new method (table 2). The two technicians making these counts had received only a few hours training in appearance and detection of mold hyphae in butter. Again the variations occurring are no greater than would be expected in a microbiological analysis.

Comparisons between values with the Wildman and new methods are too few to enable definite conclusions, but those presented in tables 3 and 4 at least indicate in a general way what might be expected. There appears to be a rough correlation between the results until the mm. per mgm. reach

TABLE 3

Comparative values with new method and Wildman method on one series of commercial butter samples

Sample No.	New method	Wildman method
	mm. per mgm.	*per cent positive fields*
66	0.88	2
296	3.26	12
294	3.64	16
295	9.72	46
293	11.53	44
297	19.29	80
298	24.89	96
299	49.54	92
300	75.01	96

about 25, after which the Wildman method shows little increase. It should be emphasized that further comparisons in large number must be made before any definite correlation between the two methods can be worked out. However, it appears from these preliminary results that when the mold content reaches about 15 mm. per mgm., a sample runs a good chance of having a Wildman count of 60, and when the mold content reaches 20 mm. per mgm., a sample will usually be definitely illegal. At about 25 mm. per mgm. the Wildman count will vary from 75–100 per cent positive fields and remain at about this figure as the mm. per mgm. increase. Commercial butters with more than 40 mm. per mgm. are not uncommon.

Two samples with the same total length of mold filament per mgm. may exhibit considerably different Wildman values. This is demonstrated best by two samples one of which contained 12.84 mm. per mgm. total mold

TABLE 4

Comparative values obtained on commercial butter samples with new method and Wildman method

Number of samples in group	New method	Average count by Wildman method	Range of Wildman counts
	mm. per mgm.	*per cent positive fields*	*per cent positive fields*
9	1– 5	10	0–16
5	5–10	33	12–46
12	10–15	49	32–68
9	15–20	69	49–80
8	20–25	80	64–96
3	25–35	87	76–96
5	35–50	93	88–96
1	75.01	96	.....

and a Wildman count of 32 and the second 12.21 mm. per mgm. and 68 per cent Wildman count. Similarly two samples with about 16 mm. per mgm. total mold, gave Wildman counts of 49 and 80 respectively.

DISCUSSION

The success in duplicating results from one sample or operator to the next emphasizes the quantitative nature of the new method. In order for the method to be actually quantitative and consistent, however, it is necessary that the operator be thoroughly familiar with microscopic appearance of mold fragments and also make absolutely certain that he is accurately estimating lengths of mold fragments in terms of mm. The ruled ocular disc employed for the Howard or Wildman counts provides sufficient rulings to enable a reasonably accurate estimation of total combined length of mold fragments in every field. Once the operator has become accustomed to estimating length of filaments, the method becomes almost as rapid as that of Wildman.

The method as it exists has some advantages and disadvantages. It does provide a truer picture of the actual mold hyphae content than any other method. The results of Claydon indicate that the degree of working in the churn affects the number and length of mold filaments in the final butter and consequently the Wildman mold mycelia value. By the new method all filaments are included whether long or short. Neither the Wildman nor the new method includes the tiniest fragments of *Oospora lactis* hyphae sometimes referred to as spores. However, examination indicated that only a comparatively small number of the tiny fragments remain in the butter. Most of them pass out into buttermilk and wash water.

There is a definite advantage to knowing the actual total mold content of a sample. The commercial samples in these studies running as high as 49 and 75 mm. per mgm. total mold filament provide an example of a problem confronting many creameries. The high temperatures and relative humidities in this part of the country are quite conducive to mold growth in cream on the farm. Creameries in this area are therefore blessed with an abundance of mold growth in the cream during the warm season. The result is that much 90 score butter runs over 60 per cent by the Wildman test. The 89 score butter is often correspondingly higher in mold content. Much of it runs as high as 90 to 100 by the Wildman test. Such butter may contain as much as 75 mm. per mgm. of mold by the new method. If the creameryman in a quality improvement program eliminates half the moldy cream, he may bring the mold content of his butter down to about 40–50 mm. Yet it still will run 90 or over by the Wildman test. If he then again eliminates 50 per cent of this moldy cream, he reduces the mold content of his butter to perhaps 25 mm. per mgm. Again instead of reducing the Wildman count 50 per cent he may end up with a 90 to 100 per cent mold mycelia count. It is not until the mold content is reduced below 20–25 mm. per mgm. that a distinct lowering in mold content of butter can be detected by the Wildman mold mycelia count. This condition has been responsible for considerable misunderstanding in cream quality improvement work.

The variation in Wildman mold mycelia count values for butters of equivalent total mold content indicates that more study should be given to factors possibly responsible for such variations. A variation of 20 or 25 per cent due possibly to manufacturing conditions may easily throw a butter from a definitely legal into an illegal class.

Another advantage of the new method is that any microscope of any field diameter can be employed as long as it provides about 100× magnification and a stage micrometer is available to determine the field diameter. Also, a permanent preparation of the sample is obtained which can be examined weeks later if more convenient. Materials other than butter can be diluted in hot gum solution and their total content of mold filament determined. Because of the interfering effect of the protein, samples of cream and butter-

milk must be diluted out further than butter. Because of its low mold content, butter wash water must be diluted less than butter. Covering the preparation with immersion oil is especially helpful in examining cream and buttermilk preparations. In cream examination, special methods are necessary to thoroughly agitate the samples in order to break up masses of mold filaments.

This method was not developed nor is it presented for the purpose of replacing the Wildman count. It may appear too technical for the average control laboratory. However, it has been about as easy to teach to a careful operator as the Wildman method. The new method, because it is quantitative, has proven a valuable research tool. Investigations on mold content of butter and other materials have been carried out that would not have been possible by any other method.

SUMMARY

A method for determination of total combined length of dead mold fragments per gram or milligram of butter has been developed.

The procedure consists of dilution of one gram of butter in hot gum solution, spreading a small quantity of this on a glass slide, staining with Newman's stain and examining with low power magnification. The average length of mold filament per field is multiplied by the microscope factor divided by 1000 and results reported in mm. per mgm. of butter.

This quantitative method has been of value in studies to determine the total mold fragment content of butter, buttermilk, wash water and cream.

ACKNOWLEDGMENT

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REFERENCES

(1) Amer. Public Health Ass'n. Standard Methods for the Examination of Dairy Products. Ed. 8, p. 53. 1941.

(2) Claydon, T. J. Some Factors Affecting the Mold Content of Cream and the Mold Mycelia Count of Butter. Ark. Agr. Expt. Sta. Bul. 432. 1943.

(3) Elliker, P. R., and Brown, W. H. Factors Affecting Mold Content and Quality of Cream. Purdue Agr. Expt. Sta. Bul. 465. 1942.

(4) Association of Official Agricultural Chemists. Methods of Analysis. Ed. 5, p. 300. 1940.

THE DETERMINATION OF CITRIC ACID IN MILK PRODUCTS BY CERATE OXIDIMETRY

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Of the various methods which have been proposed for the determination of citric acid in milk, those based on Stahre's (12) reaction have come into wide use. In this method, the citric acid is oxidized by dilute potassium permanganate in the presence of bromine resulting in the formation of insoluble pentabromacetone, which is determined gravimetrically.

Allen (1) points out three objections to this method: "1), the formation of pentabromacetone as an oil instead of a crystalline precipitate; 2), the relatively high solubility of the precipitate; 3), the loss due to volatility when moist."

By determining the solubility of the precipitate (and using a factor to include this loss in the final calculations) and by drying the precipitate in a vacuum at less than 20° C. for sixteen hours, Deysher and Holm (2) largely eliminated the second and third errors pointed out by Allen. But in so doing, the length of time of performing the test was extended another twenty-four hours, making a total length of time of three days.

If the determination of citric acid is to be used as a control test in the manufacture of dairy products, particularly evaporated milk, a much shorter method is necessary.

A search of literature, therefore, was made to ascertain if other methods would be applicable. The conductimetric method of Kolthoff (6) was tried but found to be unsuitable for milk owing to the high concentration of other ions, particularly chloride. Potentiometric titrations using silver nitrate, barium acetate, and lead acetate resulted in failures because of interfering phosphate and chloride ions. Rundell (9) states that attempts to determine citric acid potentiometrically with calcium chloride were unsuccessful.

The iodimetric method of Kometianni (7) involves precipitation of the citric acid as pentabromacetone under conditions which are not in accordance with those found necessary by Deysher and Holm for quantitative recovery. This is in agreement with Templeton's (13) statement that the iodimetric method is not quantitative. Hartman (3) also states that Kometianni's procedure is not entirely satisfactory when applied to mixed fruit acids. If the method were modified to insure accuracy, it would offer no advantage over the Deysher and Holm procedure.

Colorimetric methods in which acetic anhydride and pyridine are used to develop color directly from the citrate ion were found to be unsatisfactory, probably due to the presence of the chloride ion. Methods using

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acetone dicarboxylic acid or pentabromacetone for a basis for color development were not tried since no great saving in time would have resulted.

Willard and Young (14) showed that ceric sulphate in sulphuric acid solutions oxidized citric acid at the rate of 1.211 mg. anhydrous citric acid per 1 ml. of 0.1 N sulphato-cerate. Smith and Duke (10) found that the perchlorato-cerate ion in the presence of perchloric acid oxidized citric acid stoichiometrically at the rate of 1.372 mg. per 1 ml. of 0.1 N perchlorato-cerate. When these two methods were applied to milk, it was found that the former method was not stoichiometric. The following method is based on Hartman and Hillig's procedure (4, 5) for isolating the citric acid (omitting tartaric and phosphotungstic acids which interfere) and Smith and Duke's procedure for oxidizing it. In preliminary work, to determine the adaptability of their indicator to the method as applied to milk, a Beckman pH set was employed using a glass electrode as reference electrode and polished platinum wire as indicating electrode following the suggestion of Lykken (8).

PREPARATION OF REAGENTS

1. *Ammonium perchlorato-cerate.* Smith (11). Dissolve 55 to 56 grams of $(NH_4)_2Ce(NO_3)_6$ by adding the salt to a liter beaker containing 340 to 345 ml. of 72 per cent perchloric acid. Stir well during a half-minute interval and add 100 ml. of water. Again stir well for a half-minute interval and add an additional 100 ml. of water. Continue this procedure until the volume attains 1 liter. Transfer to a suitable glass-stoppered reagent bottle and store in the dark. It is preferable to store the perchlorato-cerate solution in a black bottle, conveniently made by completely covering a reagent bottle with black electrical insulating tape. Such a solution requires standardization about every fifteen days. Carbon or dust if allowed to come in contact with perchlorato-cerate solutions catalyze their slight deterioration rate. The perchlorato-cerate solution is standardized by measuring 20.00 ml. into a 150 ml. beaker and adding 15 ml. of 70 per cent perchloric acid, 35 ml. of water, and 1 drop of nitro-ferroin. Titrate with 0.1 N sodium oxalate to the first formation of a pink color.

2. *Sodium oxalate in 0.1 N perchloric acid.* Smith (11). Dissolve 6.706 grams of Bureau of Standards, standard of reference sodium oxalate, per liter of 0.1 N perchloric acid. This solution is stable upon storage. It is used to standardize the ammonium perchlorato-cerate solution and to titrate the excess perchlorato-cerate left after oxidation of the citric acid.

3. *Nitro-ferroin* (nitro-o-phenanthroline ferrous complex) used as internal indicator is commercially available.

4. *Lead acetate.* Dissolve 75 gms. of normal lead acetate in water, add 1 ml. of glacial acetic acid and dilute to 250 ml.

5. *Blank solution.* Dissolve 0.5 gm. lactose, 0.2 gm. $CaCl_2$, and 0.2 gm. KH_2PO_4 in 100 ml. H_2O.

6. *Saturated lead citrate.* Add a slight excess of the lead acetate solution to a 10 per cent solution of A.C.S. grade citric acid in distilled water. Wash several times with distilled water and allow to stand at least twenty-four hours. Filter through No. 42 Whatman or similar grade filter paper before using. (At 25° C., lead citrate is soluble to the extent of 6 mg. per 100 ml. of water.)

PROCEDURE

Weigh 50 grams of whole milk, 25 grams of evaporated milk, or 5 grams of milk powder in a 150-ml. beaker. Add 25 ml. of water to the evaporated milk and 45 ml. to the dried milk. Add 6 ml. of 1 N sulphuric acid and heat for fifteen minutes on a steam bath. Cool. Add 95 per cent ethyl alcohol and transfer to a 250-ml. volumetric flask. Make to 250 ml. with alcohol. Mix and filter through a good grade of filter paper (such as Whatman No. 1). If filtrate is not clear, repeat the filtration. Place 100.0 ml. of the filtrate in a 100-ml. centrifuge tube and add 4 ml. of lead acetate solution. Allow to stand 3–5 minutes with occasional shaking. Centrifuge 5 minutes. Decant, testing the alcoholic solution with a few drops of lead acetate to insure complete recovery of lead citrate. Drain for 5 minutes. Add 30 ml. of water saturated with lead citrate. Shake until precipitate is dislodged, centrifuge 5 minutes, decant, and drain for 5 minutes. Wash 2 more times with 20-ml. portions of water saturated with lead citrate, centrifuging and draining 5 minutes each time. Add 10 ml. water and shake to dislodge the precipitate. Transfer to a 250-ml. beaker using a total of 55 ml. of 70 per cent perchloric acid to wash the centrifuge tube, pouring the washings into the beaker. Place beaker in ice and salt mixture until temperature reaches – 3° to 0° C. Add 40.0 ml. 0.1 N perchlorato-cerate slowly with stirring, not allowing the temperature in the beaker to rise above 10° C. Hold for 30 minutes at 10° C. Add 95 ml. water, 2 drops of nitro-ferroin and titrate with 0.1 N sodium oxalate.

A blank determination must be run with each group as follows:

Place 25 ml. of the prepared blank solution in a 150 ml. beaker. Add 3 ml. H_2SO_4. Heat in steam bath with other samples. Cool. Add 95 per cent alcohol and make to 125 ml. Mix and filter. Continue exactly as described for milk, except that at the conclusion of the final draining period, 30 ml. of water are added and the contents of the centrifuge tube transferred to a 250-ml. beaker with 55 ml. 70 per cent perchloric acid. When the temperature has reached – 3° to 0° C., 10.00 ml. of 0.1 N perchlorato-cerate are added and the solution held for 30 minutes at 10° C. Add 95 ml. water, 2 drops of nitro-ferroin and titrate with 0.1 N oxalate. The ml. of 0.1 N cerate used constitute the blank.

$$\text{Per cent citric acid (anhydrous)} = \frac{\text{ml. 0. 1N perchlorato-cerate used} - \text{blank} \times 0.343}{\text{wt. sample}}.$$

TABLE 1

*Recovery of citric acid from pure solutions**

Citric acid taken	Recovered	Recovered
mg.	*mg.*	%
33.51	33.68	100.52
33.51	33.23	99.17
33.51	33.71	100.59
33.57	33.57	100.00
33.57	33.63	100.18
33.57	33.42	99.55

* 1 ml. 0.1 N perchlorato-cerate is equivalent to 1.372 mg. anhydrous citric acid.

The potentiometric method for the detection of the endpoint is equally as satisfactory as the use of nitro-ferroin as an internal indicator. The results included in this paper were secured with the nitro-ferroin indicator.

EXPERIMENTAL

Various quantities of anhydrous citric acid (prepared by grinding to a powder and drying Baker's analyzed and Merck's A.C.S. grade over calcium chloride) were dissolved in distilled water. Sufficient 70 per cent perchloric acid was added to make the solution 4 M with respect to the perchloric acid. After cooling the mixture to 0° C., a slight excess of 0.1 N perchlorato-cerate was added and the solution held at 10° C. for 30 minutes. It was then diluted to 2 N and titrated with 0.1 N sodium oxalate using nitro-ferroin as the indicator. The results given in table 1 show an average recovery of 100.00 per cent. It was found that if the temperature rose above 12° C., the results were high owing to side reactions as pointed out by Smith (11).

Four cultures of citric acid fermenting bacteria were secured from Iowa State College. These were *S. diacetilactis* strain N 4, and *S. citravorous* strains S 9, S 14, and Mu 29. These were inoculated into acidified sterile skim milk and incubated until citric acid was absent as shown by the pentabromacetone determination. Volumetric citrate determinations were made

TABLE 2

Recovery of citric acid from decitrated milk

Citric acid taken	Recovered	Recovered
mg.	*mg.*	%
15.1	15.1	100.0
22.2	22.8	102.7
42.4	43.3	102.1
45.4	46.0	101.3
69.3	70.0	101.0
90.0	90.1	100.1
98.9	99.0	100.1
98.9	98.8	99.9

on these cultures with known quantities of anhydrous citric acid added. These results are given in table 2. The average percentage recovery is 100.9 per cent.

It was found that decitrated milk gave a larger blank value than when the procedure was followed using only distilled water. It was thought that this might be due to some interfering substance in milk or to the formation of some interfering substance during the growth of the citric-acid-fermenting bacteria. Various natural components of milk were tested, including lactose, riboflavin, niacin, ascorbic acid, calcium and magnesium salts, phosphates, chlorides, nicotinic acid, pantothenic acid, lactic acid, propionic acid, butyric acid, acetylmethylcarbinol, and ether extract from milk but none of these was found to interfere. It was found that when the volume of the precipitate

TABLE 3

Method of preparation of blank	ml. of 0.1 N perchlorato-cerate required per 100 ml. filtrate
Fermentation of citric acid	2.35
	2.45
	3.11
	2.38
	3.27
	2.88
Precipitation of proteins and citrate with silver acetate and acetic acid	2.60
	2.94
	2.68
	1.57
Prepared blank (containing lactose, Ca, Cl, PO_4)	2.10
	2.24
	3.35
	2.87
	3.35

was the same as that obtained from milk the blank was about the same. Since repeated washings of the precipitate did not appreciably reduce the value of the blank it was thought that possibly a small amount of the lactose was occluded in the precipitation of lead citrate. However, when the thrice washed precipitate was tested for lactose using copper sulphate and alkaline tartrate solutions, no observable precipitate of copper oxide was formed. Furthermore, when perchloric acid is added to the washed precipitate of lead salts, both in the case of the decitrated milk and in the case of the various milk products examined, a yellow color is formed which does not form with any of the substances added to the blank. It is apparent, therefore, that some substances not included in this study is present in the lead salt precipitate which may interfere.

Another method for the preparation of the blank was tried, using 0.5 gm. silver acetate and 5 ml. of 10 per cent acetic acid per 50 ml. of milk in place of 6 ml. NH_2SO_4. By this procedure the citrate is precipitated as silver

citrate and filtered off. To 100 ml. of the filtrate, 4 ml. of lead acetate were added and the precipitate washed as previously described. This precipitate did not give the color reaction with perchloric acid and the blanks were slightly lower than those from the decitrated milk. However, the volume of the precipitated lead salts is small. A resume of these results is given in table 3.

Table 4 gives results of some analyses of powdered skim milk, whole milk, and evaporated milk. The volumetric citrate method gives results which are slightly higher due either to an interfering substance in milk or incomplete recovery by the pentabromacetone method.

TABLE 4

Product		Oxidation by perchlorato-cerate	Pentabromacetone (Modification of Deysher and Holm)
Powdered milk:	(1)	1.57	1.55
		1.57	1.52
	(2)	1.82	1.67
		1.87	1.63
	(3)	1.82	1.78
		1.80	1.74
Whole milk:	(1)	0.172	0.161
		0.176	0.168
	(2)	0.178	0.159
		0.168	0.167
	(3)	0.171	0.165
		0.171	0.160
Evaporated milk:	(1)	0.332	0.323
		0.325	0.327
	(2)	0.334	0.330
		0.340	0.333
	(3)	0.342	0.329
		0.333	0.338

CONCLUSIONS

A method for the determination of citric acid in milk products is presented which is based on the oxidation of citric acid (isolated from interfering substances by precipitation as lead citrate) by an excess of 0.1 N perchlorato-cerate in 4 M perchloric acid solution. The excess perchlorato-cerate is titrated with 0.1 N sodium-oxalate in 2 M perchloric acid using nitro-ferroin as an internal indicator.

Care must be used to free the citric acid from other organic substances which are also oxidized by the perchlorato-cerate. The results from the method are slightly higher than those from the pentabromacetone method and can be secured in considerably less time.

REFERENCES

(1) ALLEN, L. A. Mineral Constituents and Citric Acid Content of Milk. Jour. Dairy Res., 3: 1–51. 1931.

(2) DEYSHER, EDGAR F., AND HOLM, GEO. E. Determination of Citric Acid in Pure Solutions and in Milk by the Pentabromacetone Method. Indus. and Engin. Chem., Analyt. Ed., **14**: 4–7. 1942.

(3) HARTMAN, B. G. Report on Fruit Acids. Assoc. Off. Agr. Chem. Jour., **24**: 386–391. 1941.

(4) HARTMAN, B. G., AND HILLIG, F. Application of the Stahre Reaction to the Accurate Determination of Citric Acid. Assoc. Off. Agr. Chem. Jour., **10**: 264–272. 1927.

(5) HARTMAN, B. G., AND HILLIG, F. Citric Acid in Milk. Assoc. Off. Agr. Chem. Jour., **15**: 643–645. 1932.

(6) KOLTHOFF, I. M. The Use of Conductivity Titrations in Precipitation Analysis. V. Conductrimetric Titrations with Barium Salt. Ztschr. f. Analyt. Chem., **61**: 433–438. 1922.

(7) KOMETIANNI, P. A. A New Iodimetric Method for Determining Citric Acid. Ztschr. f. Analyt. Chem., **86**: 359–366. 1931.

(8) LYKKEN, LOUIS, AND TUEMMLER, L. D. Glass Electrode as a Reference Electrode in Electrometric Titrations. Indus. and Engin. Chem., Analyt. Ed., **14**: 67–69. 1942.

(9) RUNDELL, K. Determination of Citric Acid in Food Products. Soc. Chem. Indus. Victoria Proc., **39**: 183–188. 1939. (See C. A., **34**: 2078.)

(10) SMITH, G. FREDERICK, AND DUKE, F. R. Cerate and Periodate Oxidimetry. Indus. and Engin. Chem., Analyt. Ed., **15**: 120–122. 1943.

(11) SMITH, G. FREDERICK. Cerate Oxidimetry. G. Frederick Smith Chemical Company. Columbus, Ohio. 1942.

(12) STAHRE, L. Nord Pharm. Tids, **2**: 141. 1895. (Original not seen. See reference (4) or LAMPITT, L. H., AND BOGOD, M. Chim. and Indus., **31**: 777. 1931.)

(13) TEMPLETON, HUGH L. Determination of Citric Acid in Milk. JOUR. DAIRY SCI., **19**: 472. 1936.

(14) WILLARD, H. H., AND YOUNG, PHILENA. Ceric Sulphate as a Volumetric Oxidizing Agent. Jour. Amer. Chem. Soc., **52**: 132–142. 1930.

THE EFFECT OF IODINATED CASEIN (PROTAMONE) ON MILK AND BUTTERFAT PRODUCTION AND ON THE ASCORBIC ACID CONTENT OF THE MILK*

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In 1934, Graham (3) reported the results of extensive studies with thyroidectomized dairy cattle and with the feeding of desiccated thyroid glands to lactating cows. In this experiment the diminution in milk secretion following the removal of the thyroid could not be distinguished readily from that accompanying a control operation without the removal of the thyroid gland; however, there was a distinct rise in milk secretion, when small amounts of dried thyroid glands were fed either to thyroidectomized cows or to unoperated normal cows in the declining phases of lactation. The stimulating factor in the thyroid glands was shown to be thyroxine by an increase in milk secretion when synthetic thyroxine was injected (4). These observations have since been confirmed and extended by Jack and Bechdel (7), Folley and White (2), and Herman, Graham and Turner (5).

The practical value of feeding pure thyroxine or desiccated thyroid glands to increase milk production was largely nullified by the exorbitant cost of this material. In 1940, Turner (13) reported a possible cheap source of material with thyroxine activity in the form of iodinated casein. Reineke and Turner (10) have recently described a method for the preparation and the use of this synthetic thyroprotein. This material is prepared by the iodination of casein in solution to which sufficient sodium bicarbonate has been added to maintain the pH of the reaction mixture within the range of 6.8 to 8.0.

The administration of this artificial thyroprotein to cows and goats, according to Reineke and Turner (11), stimulates increases in milk production and the percentage of fat in the milk similar in every respect to those produced by either desiccated thyroid glands or synthetic thyroxine.

The purpose of this paper is to report the results obtained with lactating dairy cows in the declining part of the lactation period, when the iodinated casein was fed in smaller quantities but for longer periods of time than that reported previously by Reineke and Turner (11).

EXPERIMENTAL

Six pure-bred Holstein cows in the sixth to the eighth month of their first lactation were selected from the regular milking herd for this experiment.

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The cows were producing from 30 to 40 pounds of milk per day at the beginning of the experiment. The experiment was a double reversal feeding trial started on the twenty-first day of February and continued for a period of 13 weeks or until May 22. The first week was used as an adjustment period at which time the cows were divided into two equal groups of three cows each on the basis of milk production, fat production, and body weight.

Group 1, composed of cows number 535, 536, and 544 was fed 15 grams of iodinated casein (Protamone) per cow daily as a supplement to the regular ration for the first 4-week period. The iodinated casein was discontinued for the second 4-week period after which it was again fed during the third 4-week period.

Group 2, consisting of cows number 527, 529, and 541, served as a check for the first 4-week period and then was fed 15 grams of the iodinated casein per cow daily for the second 4-week period, after which it was discontinued for the last or the third 4-week period.

During the first two 4-week periods, the cows were confined to the barn except when they were allowed to run outside in a clean lot for exercise. During the last 4-week period of the experiment the cows were on pasture. The ration fed the cows in each group was composed of a concentrate mixture, legume hay and beet pulp. The cows were fed daily 10 pounds of concentrates, 2 pounds beet pulp and all the hay they would clean up. They were fed a uniform amount of feed throughout the experiment regardless of the amount of milk produced and the gain or loss in body weight.

The cows were milked twice daily. On Monday, Wednesday, and Friday of each week they were weighed and one-half pint samples of milk were collected night and morning for analysis. Each sample of milk was analyzed for fat by the Babcock method and for total solids by the oven method. Ascorbic acid was titrated in the samples collected each morning with 2–6 dichlorophenolindophenol as described by Sharp (12).

Close observations were made each day for any unusual reaction which might become apparent. A maximum-minimum thermometer placed in the barn was read daily in order to note any unusual change in temperature. Respiration and pulse rates were obtained between 2 and 4 P.M. on Friday of each week during the first two 4-week periods when the cows were confined to the barn. The pulse was counted by feeling the posterior tibial artery on the medial surface of the tibia at a point eight to ten inches above the hock joint.

RESULTS

Data showing the effect of feeding iodinated casein on milk and butterfat production; the percentage fat and solids-not-fat; the ascorbic acid in the milk and changes in body weight are presented in table 1 and in figures 1 to 6. Data for individual days were averaged and presented as weekly averages for individual animals or as groups as the case might be. The

figures represent averages for the separate groups except in the case of figure 4, which shows the effect of iodinated casein on solids-not-fat and where variation in the results obtained made it seem desirable to present data for individual animals. It may be observed in table 1, that data for 529 are incomplete. This animal appeared normal until about the middle of the second 4-week feeding period. She had received 15 grams of the iodinated casein daily for two weeks when she began to show the symptoms of a rabid animal. She refused to eat and lost in body weight rapidly, and died in about a week after showing the first symptoms. After an examination of the brain, the cause of the death of the animal was definitely diagnosed as rabies. Data from this animal are not included in the averages for Group 2.

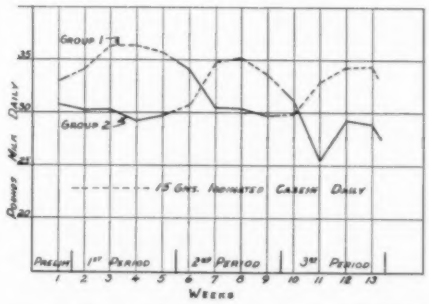

FIG. 1. Effect of iodinated casein on milk production.

Effect on milk production. Data presented in table 1 and in figure 1 show that the feeding of 15 grams of iodinated casein daily, as a supplement to the regular ration, did cause a definite increase in the milk production of all animals on experiment. It may be observed in figure 1 that the cows increased in milk production for about two or three weeks after the beginning of iodinated casein feeding, after which there was a tendency to show a slight downward trend. It may be observed in the case of the cows in Group 2 that milk production showed a sharp decline for about two weeks during the third period when iodinated casein had been discontinued. During the latter part of the third period there was an increase in milk production which is thought to be due to the cows going out on pasture. At that particular time the cows in Group 1 were receiving iodinated casein which probably supplemented the stimulating effect of pasture.

Effect on percentage fat content of the milk. Data summarized in figure 2, show the effect of feeding iodinated casein on the fat content of the milk. The cows in both groups showed an increase in percentage fat content of the milk during the feeding periods. It may be observed that there was considerable carry-over effect following the feeding of the iodinated casein. After the feeding of the iodinated casein, the cows continued to secrete milk

TABLE 1

Average daily milk and fat production, the percentage fat, and solids-not-fat and the ascorbic acid content of milk

Weeks	Date	Milk, lbs.	Fat, %	Fat, lbs.	S-N-F, %	Ascorbic acid, mg./liter	Milk, lbs.	Fat, %	Fat, lbs.	S-N-F, %	Ascorbic acid, mg./liter	Milk, lbs.	Fat, %	Fat, lbs.	S-N-F, %	Ascorbic acid, mg./liter
		Group I														
		Cow Number 535					Cow Number 536					Cow Number 544				
1	Feb. 21–27	37.0	3.83	1.42	8.53	20.2	32.8	4.07	1.33	9.00	18.4	29.1	3.62	1.05	8.41	16.7
	15 grams of iodinated casein daily															
2	Feb. 28–Mar. 6	35.8	3.81	1.36	8.50	19.2	34.6	4.13	1.43	9.03	15.3	32.0	3.83	1.23	8.56	14.4
3	Mar. 7–13	38.8	4.07	1.58	8.87	16.2	36.8	4.42	1.63	9.00	12.9	33.7	3.99	1.34	8.78	19.9
4	Mar. 14–20	39.0	4.30	1.68	8.77	15.2	36.0	4.51	1.62	8.99	11.9	34.2	4.20	1.44	8.78	9.9
5	Mar. 21–27	38.7	4.22	1.63	8.75	15.7	35.7	4.68	1.67	8.92	12.3	33.0	4.41	1.46	8.69	11.5
	Iodinated casein discontinued															
6	Mar. 28–Apr. 3	37.0	4.14	1.53	8.93	17.8	33.3	4.68	1.56	9.12	14.3	31.9	4.11	1.31	8.74	14.2
7	Apr. 4–10	34.3	4.58	1.57	8.50	17.6	28.6	4.65	1.33	9.12	15.9	28.7	4.07	1.17	8.94	15.4
8	Apr. 11–17	34.2	3.79	1.30	8.64	16.2	29.5	4.81	1.42	9.10	13.4	27.5	3.95	1.09	8.80	13.0
9	Apr. 18–24	31.6	4.15	1.31	8.43	17.3	28.9	4.41	1.27	9.23	15.3	28.9	3.90	1.13	8.74	15.1
	15 grams of iodinated casein daily, and cows on pasture															
10	Apr. 25–May 1	32.1	4.00	1.28	8.56	15.7	28.1	4.39	1.23	9.15	13.6	29.4	3.93	1.16	8.71	10.7
11	May 2– 8	34.7	4.55	1.58	8.43	12.9	33.4	4.67	1.56	9.07	10.7	30.2	4.48	1.35	8.67	8.8
12	May 9–15	37.0	4.73	1.75	8.35	13.9	34.9	6.10	2.13	9.19	11.3	30.9	5.10	1.58	8.57	9.3
13	May 16–22	37.4	4.59	1.72	8.34	15.7	32.3	5.37	1.73	9.25	12.4	33.2	4.58	1.52	8.76	10.9

TABLE 1—(*Continued*)

Weeks	Date	Milk, lbs.	Fat, %	Fat, lbs.	S–N–F, %	Ascorbic acid, mg./liter	Milk, lbs.	Fat, %	Fat, lbs.	S–N–F, %	Ascorbic acid, mg./liter	Milk, lbs.	Fat, %	Fat, lbs.	S–N–F, %	Ascorbic acid, mg./liter
Group II																
		Cow Number 527					Cow Number 529					Cow Number 541				
1	Feb. 21–27	34.6	3.77	1.30	8.36	17.2	33.0	3.87	1.28	8.53	19.7	27.0	3.25	0.88	8.51	19.0
2	Feb. 28–Mar. 6	34.4	3.88	1.33	8.30	17.0	31.1	3.78	1.18	8.63	19.5	25.9	3.76	0.97	8.41	19.9
3	Mar. 7–13	34.4	3.77	1.30	8.33	17.7	32.9	3.77	1.24	8.62	19.4	26.1	3.52	0.92	8.56	19.3
4	Mar. 14–20	32.6	3.83	1.25	8.15	15.9	31.1	3.98	1.24	8.60	19.0	25.9	3.52	0.91	8.45	18.0
5	Mar. 21–27	33.8	3.85	1.30	8.25	15.8	30.2	4.03	1.22	8.69	18.7	25.7	3.67	0.94	8.55	17.7
	15 grams of iodinated casein daily															
6	Mar. 28–Apr. 3	34.4	3.81	1.31	8.17	12.7	32.9	3.69	1.21	8.68	11.9	27.0	3.46	0.93	8.72	16.6
7	Apr. 4–10	38.6	4.32	1.67	8.39	9.6	35.3	3.50	1.24	8.77	13.8	31.1	3.60	1.12	8.64	12.1
8	Apr. 11–17	39.1	4.21	1.65	8.99	8.6	.....*	.....	.....	.....	.....	31.3	3.97	1.24	9.10	12.2
9	Apr. 18–24	36.3	4.19	1.52	8.76	10.0	.....	.....	.....	.....	.....	31.1	4.52	1.41	8.89	13.5
	Iodinated casein discontinued, cows on pasture															
10	Apr. 25–May 1	32.8	4.12	1.35	8.89	13.3	.....	.....	.....	.....	.....	29.4	3.93	1.16	8.94	13.2
11	May 2– 8	27.3	4.64	1.27	8.67	15.5	.....	.....	.....	.....	.....	23.3	4.34	1.01	8.81	16.2
12	May 9–15	32.4	4.32	1.40	8.35	13.1	.....	.....	.....	.....	.....	26.2	4.71	1.23	8.39	14.4
13	May 16–22	31.7	4.06	1.29	8.39	14.1	.....	.....	.....	.....	.....	26.2	4.28	1.12	8.33	12.8

* Cow died with rabies.

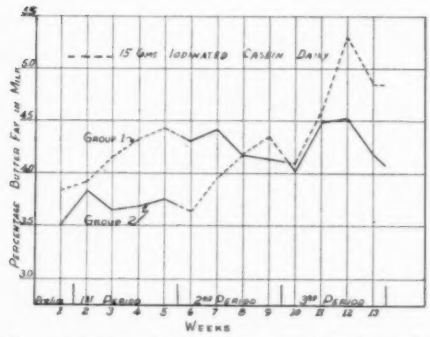

FIG. 2. Effect of iodinated casein on percentage butterfat in milk.

of a higher fat content than before, for a period of at least four weeks. In the case of the cows in Group 1, there was an increase from an average of about 3.8 per cent to about 4.4 at the end of the first 4-week feeding period. During the next 4-week period when the cows did not receive the iodinated casein the percentage fat decreased only about 0.2 per cent. During the last 4-week feeding period when the cows were again fed iodinated casein, the fat percentage increased to about 5 per cent. This represents about 1.2 per cent increase in the fat content of the milk during the experiment. A small part of this increase in fat percentage may have been due to advance in lactation.

The cows in Group 2 showed an increase in the percentage fat from about 3.7 to about 4.4 per cent after four weeks of casein feeding. They were able to maintain a level of about 4.4 per cent for the following four weeks when they received no iodinated casein.

Effect on total fat production. Data summarized in figure 3, show the effect of iodinated casein on total fat production. In general butterfat pro-

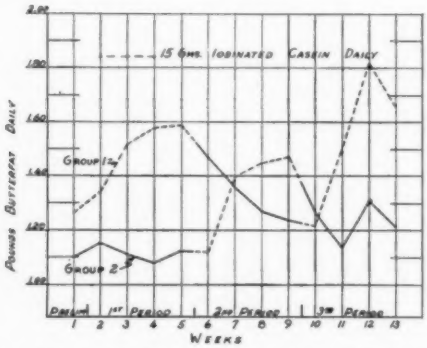

FIG. 3. Effect of iodinated casein on butterfat production.

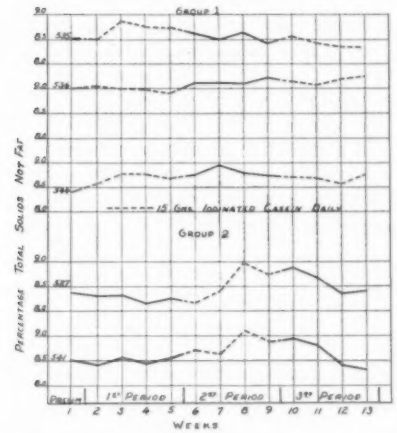

FIG. 4. Effect of iodinated casein on the percentage total solids-not-fat of milk.

duction followed about the same trend as milk production for the animals in both groups. The relative change in fat production due to casein feeding was considerably greater than in the case of milk production. This was brought about by both an increase in milk production and an increase in the percentage fat content of the milk.

Effect on the solids-not-fat content of the milk. In table 1 and in figure 4 are presented data for the solids-not-fat content of the milk for the animals in both groups. It may be observed that the changes in the solids-not-fat content are small, but there seems to be a tendency for a small increase for animals Number 535 and 544 during the first iodinated casein feeding periods, with a slight decrease during the next period for Number 535 when she did not receive iodinated casein. In the case of cows Number 527 and

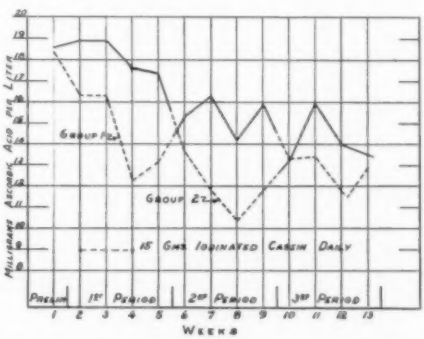

FIG. 5. Effect of iodinated casein on ascorbic acid content of milk.

541, there seems to be a much more definite tendency for the solids-not-fat to show an increase during the iodinated casein feeding with a decrease during the next 4-week period when the cows did not receive any iodinated casein.

Effect on the ascorbic acid content of the milk. In figure 5 is presented a summary of the data persented in table 1, showing the effect of feeding iodinated casein on the ascorbic acid content of the milk. It may be observed that there was a definite decrease in the ascorbic acid content of the milk in all cases when iodinated casein was fed. When the iodinated casein feeding was discontinued there was an increase in the ascorbic acid in the milk, but there seemed to be some carry-over effect. It may be observed that the general trend was downward as the experiment progressed.

Effect on change in body weight of the cows. In figure 6 is presented a summary of the changes in body weight of the cows in both groups. It may

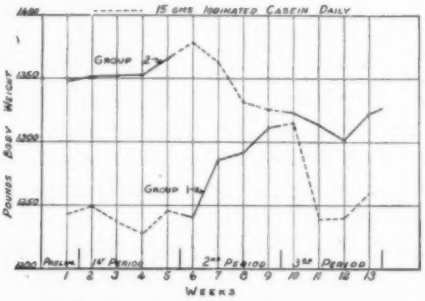

FIG. 6. Effect of iodinated casein on body weight.

be noted that the cows in Group 1 did not show any particular change in body weight during the first iodinated casein feeding period but showed an increase of about seventy pounds during the second 4-week period when iodinated casein was not fed. During the third period when they received iodinated casein they showed a decrease in body weight which was about equal to the gain made during the preceding period.

The cows in Group 2 showed a tendency to gain slightly in body weight during the first 4-week period when not receiving iodinated casein but declined considerably during the second 4-week period when they received iodinated casein. They continued to show a decline in body weight for about two weeks during the third period when they were not receiving iodinated casein. After the middle of the third period body weight began showing an upward trend when they were turned to pasture.

Effect on pulse rate and respiration. In table 2 are presented data showing the effect of iodinated casein on the pulse and respiration rates. Data are also presented showing the minimum and maximum barn temperatures on the days that the pulse and respiration rates were determined.

TABLE 2

Effect of iodinated casein on the pulse rate and respiration

	Group 1								Group 2								Barn temperature, °F.	
	535		536		544		Average		527		529		541		Average			
	Pulse	Resp.	Pulse	Resp.	Pulse	Resp.	Pulse	Resp.	Pulse	Resp.	Pulse	Resp.	Pulse	Resp.	Pulse	Resp.	Min.	Max.
Feb. 23	69	22	70	26	68	28	69	25	65	25	62	26	68	24	65	25	58	67
Feb. 26	62	18	69	16	57	18	63	17	71	16	58	20	57	16	62	17	40	46
15 grams iodinated casein daily																		
Mar. 5	72	20	80	24	68	21	73	22	62	17	65	16	63	22	64	18	42	54
Mar. 11	76	28	80	30	68	28	75	29	60	15	64	17	62	20	62	17	45	57
Mar. 20	76	22	72	20	74	25	74	22	64	18	65	16	64	22	64	19	48	54
Mar. 26	76	22	84	30	84	33	81	28	62	22	60	22	64	28	62	24	54	65
Iodinated casein discontinued									15 grams iodinated casein daily									
Apr. 2	64	18	72	25	76	26	71	23	72	26	80	33	68	27	73	29	43	66
Apr. 9	64	24	64	32	72	36	67	31	80	32	68	25	72	24	73	27	54	70
Apr. 16	64	18	60	22	72	35	65	25	72	27	....†	.....	68	24	68	25	44	60
Apr. 23	60	28	64	25	68	28	64	27	80	35	.....	.....	72	36	76	36	55	71
15 grams iodinated casein daily									Iodinated casein discontinued									
Apr. 30	64	25	72	30	72	42	69	32	64	27	.....	.....	64	25	64	26	43	72
May 7*	80	50	88	60	72	50	80	53	72	24	.....	.....	64	40	68	32	60	80

* Cows were on pasture during forenoon.

† Animal died with rabies Data not included in average for group.

It may be observed that there was considerable variation in the pulse rate and respiration rate for the cows in the two groups, nevertheless there was a definite tendency for an increase in pulse rate and respiration with iodinated casein feeding. There was also an increase in respiration with an increase in the barn temperature as might be expected.

DISCUSSION

The results obtained in this experiment show clearly that iodinated casein will stimulate an increase in milk production and the percentage fat in the milk. There is some indication of a slight increase in the percentage solids-not-fat and a considerable decrease in the ascorbic acid content of the milk.

The effect on milk production and the composition of the milk was observed after about 4 to 5 days of iodinated casein feeding. Reineke and Turner (11) observed an increase in milk production after the third day when they were feeding from 50 to 100 grams daily. Cows in the declining phase of lactation fed 15 grams of iodinated casein daily (7.5 grams morning and evening) for a period of four weeks showed an increase of from 5 to 20 per cent in milk production and from 25 to 50 per cent in butterfat production.

The increase in butter fat production was due to both an increase in milk production and an increase in the percentage fat content of the milk. There was considerable carry-over effect from one 4-week period to another in the percentage fat content of the milk. There is some evidence that the cows did not reach the peak for fat percentage of the milk in the 4-week feeding periods. It may be seen that the cows in Group 1 continued to rise in the percentage fat of the milk for the second 4-week iodinated casein feeding period. The butter fat percentage appeared to be influenced more by iodinated casein feeding than milk production.

The solids-not-fat content of the milk of cows fed iodinated casein was only slightly increased which was probably not significant. Herman, Graham and Turner (6) obtained only a slight increase in the solids-not-fat content of the milk when thyroxine was injected subcutaneously or when desiccated thyroid was fed orally.

The feeding of 15 grams of iodinated casein daily was sufficient to cause a decrease of approximately thirty-three per cent in the ascorbic acid content of the milk. This is very similar to results reported earlier by Brown, Van Landingham and Weakley (1) when potassium iodide was fed. When fed at the rate of 5 grams daily for two weeks postassium iodide did not cause an increase in the rate of milk production. What effect, if any, iodinated casein may have on the ascorbic acid content of the blood is yet to be determined. Nevertheless one might expect a reduction in the ascorbic acid content of the blood corresponding to the reduction in the ascorbic acid content

of the milk. Recent investigations have shown that the level of ascorbic acid in the blood plasma is closely associated with fertility in dairy cattle. Results obtained by Philips *et al.* (8, 9) with ascorbacidotherapy on sterile bulls and "hard to settle" cows indicate that ascorbic acid is an important factor in breeding efficiency.

Data obtained on the effect of feeding iodinated casein on respiration and pulse rate are very limited, nevertheless there was a tendency for the rate of respiration to increase. There was also an increase of about 10 beats per minute in pulse rate when iodinated casein was fed. This seems to be in line with observations of Reineke and Turner (11).

Changes in body weight were noted but probably did not affect the animal adversely in the short time covered by this experiment.

SUMMARY

The effect of feeding 15 grams of iodinated casein (Protamone) to milking cows in the declining part of their first lactation has been studied and the following results obtained:

1. Changes in milk production and in the composition of the milk were apparent after about four to five days of iodinated casein feeding.

2. Cows in the declining part of lactation showed an increase of 5 to 20 per cent in milk production and from 25 to 50 per cent in butterfat production.

3. During the first four-weeks' feeding of iodinated casein the fat content of the milk was increased by 0.47 to 0.98 per cent above the fat content of the milk at the beginning of the experiment. When iodinated casein was discontinued for four weeks and then fed for a second four-week period, the fat content was increased by 0.90 to 2.03 per cent above the fat content of the milk at the beginning of the experiment.

4. There was only a slight increase in the solids-not-fat content of the milk.

5. There was a decrease of about thirty-three per cent in ascorbic acid content of the milk.

6. There was also an increase in respiration and pulse rate, and a small decrease in body weight.

ACKNOWLEDGMENT

The authors are indebted to Dr. W. Carson Brown of the Department of Dairy Husbandry and to Dr. V. B. Fish of the Department of Agricultural Chemistry for their assistance with the analytical work in connection with this study. The authors are also indebted to Dr. W. R. Graham, Jr., of the Cerophyl Laboratories, Inc., Kansas City, for supplying the iodinated casein (Protamone) used in these trials.

REFERENCES

(1) Brown, W. Carson, Van Landingham, A. H., and Weakley, Jr., Chas. E. Oxidized Flavor in Milk. X. The Effect of Feeding Potassium Iodide Supplements to Dairy Cows on the Carotene Content of the Butter Fat and on the Ascorbic Acid Content of the Milk and the Relationship to Metal-induced Oxidized Flavor. Jour. Dairy Sci., 24: 1035. 1941.

(2) Folley, S. J., and White, P. The Effect of Thyroxine on Milk Secretion and on the Phosphatase of the Blood and Milk of the Lactating Cow. Proc. Royal Soc., London, Series B, 120: 346. 1936.

(3) Graham, Jr., W. R. The Effect of Thyroidectomy and Thyroid Feeding on the Milk Secretion and Milk Fat Production of Cows. Jour. Nutr., 7: 407. 1934.

(4) Graham, Jr., W. R. The Action of Thyroxine on the Milk and Milk Fat Production of Cows. Biochem. Jour., 28: 1368. 1934.

(5) Herman, H. A., Graham, Jr., W. R., and Turner, C. W. The Effect of Thyroxine on Milk and Fat Production. Jour. Dairy Sci., 20: 412. 1937.

(6) Herman, H. A., Graham, Jr., W. R., and Turner, C. W. The Effect of Thyroid and Thyroxine on Milk Secretion in Dairy Cattle. Mo. Agr. Expt. Sta. Res. Bul. 275. 1938.

(7) Jack, E. L., and Bechdel, S. I. A Study of the Influence of Thyroxine on Milk Secretion. Jour. Dairy Sci., 18: 195. 1935.

(8) Philips, Paul H., Lardy, H. A., Boyer, P. D., and Werner, George M. The Relationship of Ascorbic Acid to Reproduction in the Cow. Jour. Dairy Sci., 24: 153. 1941.

(9) Philips, Paul H., Lardy, H. A., Heizer, E. E., and Rupel, I. W. Sperm Stimulation in the Bull Through the Subcutaneous Administration of Ascorbic Acid. Jour. Dairy Sci., 23: 873. 1940.

(10) Reineke, E. P., and Turner, C. W. Formation in Vitro of Highly Active Thyroproteins, Their Biologic Assay, and Practical Use. Mo. Agr. Expt. Sta. Res. Bul. 355. 1942.

(11) Reineke, E. P., and Turner, C. W. Increased Milk and Fat Production Following the Feeding of Artificially Formed Thyroproteins (Thyrolactin). Jour. Dairy Sci., 25: 393. 1942.

(12) Sharp, Paul F. Rapid Method for the Quantitative Determination of Reduced Ascorbic Acid in Milk. Jour. Dairy Sci., 21: 85. 1938.

(13) Turner, C. W. Thyrolactin, a New Source of Thyroxine for Dairy Cattle. Jour. Dairy Sci., 23: 535. 1940.

PROGRAM

THIRTY-NINTH ANNUAL MEETING

OF THE

AMERICAN DAIRY SCIENCE ASSOCIATION

OHIO STATE UNIVERSITY

COLUMBUS, OHIO

JUNE 20–22, 1944

PROGRAM COMMITTEE

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H. P. DAVIS, *Chairman*—University of Nebraska
DWIGHT ESPE—Iowa State College
O. F. GARRETT—M & R Dietetic Laboratories
E. C. SCHEIDENHELM—Michigan State College
L. H. BURGWALD—Ohio State University

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REGISTRATION

CAMPBELL HALL

OHIO STATE UNIVERSITY

Meetings will be held in buildings on the campus of Ohio State University and headquarters will be there.

COMMITTEE MEETINGS

Suitable rooms will be available for all committees and for other groups which may desire to meet. L. H. Burgwald will have charge of room assignments.

SCHEDULE OF PROGRAMS

(Eastern War Time)

Date and Time	General	Production Section	Extension Section	Manufacturing Section
Tuesday				
June 20, 1944				
9: 00–12: 00	Opening Session			
1: 00– 4: 00		Section A Section B	Section	Section A Section B
4: 00– 5: 00	Committees	Committees	Committees	Committees
7: 00	Informal Get Together	Symposium		
Wednesday				
June 21, 1944				
9: 00–11: 00		Section A Section B	Section	Section A Section B
11: 00–12: 00		Joint Business Meeting		Business Meeting
1: 00– 4: 00	Post War Problems in Dairying			
4: 00– 5: 00	Committees	Committees	Committees	Committees
7: 00	Reception			
Thursday				
June 22, 1944				
9: 00–11: 00		Joint Symposium		Symposium
11: 00–12: 00		Business Meeting	Business Meeting	Business Meeting
1: 00– 3: 30	Latin American Dairying			
3: 30	Business Meeting			
7: 00	Association Banquet			

PHOTOGRAPHIC PROJECTION

Projection equipment will be available if required. It is the hope of the committee, however, that tabular and graphic material will be presented in mimeograph form rather than as slides.

INVITATION TO VISIT OHIO EXPERIMENT STATION

The administration of the Ohio Experiment Station and the members of the Dairy Department staff cordially invite you to visit the Experiment Station at Wooster on your way to or from the Dairy Science meetings. In addition to the regular experimental herd, there is a special herd and farm devoted entirely to pasture research, an experimental forage harvester, and extensive batteries of experimental silos.

Wooster is 90 miles north of Columbus on state routes 3, 5, and 30. It is readily accessible by bus from all directions and by rail from east and west.

GENERAL PROGRAM

(Eastern War Time)

Tuesday, June 20, 1944

9:00–12:00 OPENING SESSION. Campbell Hall, *Room 200.*

A. C. DAHLBERG, *President, American Dairy Science Association*, Presiding.

Introduction of Officers and Guests.

Address of Welcome—DR. HOWARD L. BEVIS, *President, The Ohio State University.*

Response and Presidential Address—A. C. DAHLBERG, *President, American Dairy Science Association.*

Guest Speaker.

Announcements.

1:00– 4:00 SECTIONAL MEETINGS.

Production Section A, Feeds and Feeding, *Room 100, B & Z Bldg.*

Production Section B, Physiology and Nutrition, *Room 209, B & Z Bldg.*

Manufacturing Section A, Bacteriology, *Room 302, Campbell Hall.*

Manufacturing Section B, Chemistry, *Room 200, Campbell Hall.*

Extension Section, D.H.I.A. work, *Room 206, H & F Bldg.*

4:00– 5:00 COMMITTEE MEETINGS.

7:00– 9:00 **Production Section Symposium,** *Room 100, B & Z Bldg.*

Relation of Body Size to Milk Yield.

A. The Energy-Size Basis of Measuring Milk Yield—W. L. GAINES, *University of Illinois.*

B. The Significance of Body Size in Milk Production—SAMUEL BRODY, *University of Missouri.*

C. Pertinent Data on the Problem—MAX KLEIBER, *University of California.*

D. How the Size Problem Affects Breed Associations—C. T. CONKLIN, *Ayrshire Breeders Association.*

7:00 INFORMAL GET TOGETHER—*Faculty Club, Ohio State University.*

Wednesday, June 21, 1944

9:00–11:00 SECTIONAL MEETINGS.

Production Section A, Feeds and Feeding, *Room 100, B & Z Bldg.*

Production Section B, Animal Breeding, *Room 209, B & Z Bldg.*

Manufacturing Section A, Butter and Cheese, *Room 203, Campbell Hall.*

Manufacturing Section B, Milk and Other Dairy Products, *Room 200, Campbell Hall.*

Extension Section, *Room 206, H & F Bldg.*

11:00–12:00 BUSINESS MEETINGS OF SECTIONS.

Production and Extension, Joint Business Meeting, *Room 206, H & F Bldg.*

Manufacturing, Business Meeting, *Room 200, Campbell Hall.*

1:00– 4:00 GENERAL SESSION.

Post War Problems of Dairying, *Room 200, Campbell Hall.*

A. Why Different Problems After the War—E. L. ANTHONY, *Michigan State College.*

B. Is Dairy Expansion to Continue?—EARL WEAVER, *Michigan State College.*

C. Improving a Post War Testing Program—C. T. CONKLIN, *Ayrshire Breeders' Association.*

D. Post War Demand for Livestock—O. E. REED, *Bureau of Dairy Industry.*

E. Recommended Nutrient Allowances for Dairy Animals—J. K. LOOSLI, *Cornell University.*

F. Nutrition Education as a Safeguard Against Post War Problems—E. M. HARMON, *National Dairy Council.*

G. Problems of Farm Milk Collection Delivery to Plants and Retail Delivery of Milk—LELAND SPENCER, *Cornell University.*

H. Problems Pertaining to Processing, Manufacturing, and Distribution of Dairy Products in the Post War Period—H. H. SOMMER, LELAND SPENCER, A. W. FARRALL AND R. J. RAMSEY. *A Committee Report.*

4:00– 5:00 COMMITTEE MEETINGS.

7:00 RECEPTION, *Faculty Club.*

Thursday, June 22, 1944

9:00–11:00 SECTIONAL MEETINGS.

Manufacturing Section Symposium, *Room 200, Campbell Hall.*

Dehydrated Milk and Milk Products.

A. Types of Dehydrated Milk and Milk Products—P. H. TRACY, *University of Illinois.*

B. Standard Specifications and Methods of Analysis of Dried Milk—Lt. Robert Remaley, *Subsistence Research and Development Laboratory, U. S. Army Quartermaster Corps.*

C. Use of Anti-oxidants in the Manufacture and Storage of Dried Whole Milk—S. T. Coulter, *University of Minnesota.*

D. Packaging of Dehydrated Milk and Milk Products—Major James d'A. Clark, *Subsistence Research and Development Laboratory, U. S. Army Quartermaster Corps.*

E. The Future of the Dehydrated Milk and Milk Products Industry—J. T. Walsh, *American Dry Milk Institute.*

Production and Extension Symposium, *Room 206, H & F Bldg.*

Mastitis and the Feed Situation.

A. Mastitis from the Dairyman's Stand Point—T. S. Sutton, *Ohio State University.*

B. Modern Methods of Treating Mastitis—C. S. Bryan, *Michigan State College.*

Discussion

C. The Feed Situation—Walter Berger, *Chairman of the Feed and Livestock Division, Food Production Administration.*

Discussion

11:00–12:00 **Sectional Business Meetings**—Production, *Room 100, B & Z Bldg.;* Manufacturing, *Room 200, Campbell Hall;* and Extension, *Room 206, H & F Bldg.*

1:00– 3:30 **General Session,** *Room 200, Campbell Hall.*

Latin American Dairying.

A. Dairying and Agriculture—R. E. Hodgson, *Bureau of Dairy Industry.*

B. Dairy Products in Latin America—A. C. Dahlberg, *Cornell University.*

C. Thomas Jefferson, The Farmer—O. E. Reed, *Bureau of Dairy Industry.*

3:30 **General Business Meeting,** *Room 200, Campbell Hall.*

7:00 **Annual Banquet—Installation of Officers and Presentation of Borden Awards.**

SECTIONAL PROGRAMS

EXTENSION SECTION

Tuesday, June 20

Afternoon Session, Room 206, H & F Bldg.
E. C. SCHEIDENHELM—*Chairman*

1:00– 1:15 **Opening Business Session.**

1:15– 4:00 **Sectional Program.**

A. Panel Discussion on D.H.I.A. work—*Leader*, E. H. LOVELAND, *University of Vermont.*

E1. Agreement Forms and D.H.I.A. Organization —W. T. CRANDALL, *Cornell University.*

E2. Short Cuts in Record Keeping—J. F. KENDRICK, *Bureau of Dairy Industry.*

E3. A Suggested Form for Permanent and Current Records—J. F. KENDRICK, *Bureau of Dairy Industry.*

E4. Securing Supervisors During and Immediately Following the War—C. R. GEARHART, *Pennsylvania State College.*

E5. Streamlined Testing for Everybody—G. W. VERGERONT, *University of Wisconsin.*

E6. Testing in Areas of Small Herds and Scarce Membership—R. A. CAVE, *South Dakota State College.*

B. **Report of the Committee**—C. R. GEARHART, *Pennsylvania State College.*

E7. Wartime 4–H Dairy Club Program—J. C. NAGEOTTE, *Pennsylvania State College.*

C. **Report of 4–H Club Committee**—H. A. WILLMAN, *Cornell University.*

4:00– 5:00 **Committee Meetings.**

Wednesday, June 21

Morning Session, Room 206, H & F Bldg.
E. C. SCHEIDENHELM—*Chairman*

9:00–11:00 **Sectional Program.**

E8. The Forced Ventilation System of Curing Hay in the Mow—R. G. CONNELLY—*Virginia Polytechnic Institute.*

E9. A Complete Sire Proving Program—C. T. CONKLIN, *Ayrshire Breeders Association.*

E10. The Task of Getting Quality Bulls Giving Quality Semen—S. J. BROWNELL, *Cornell University.*

E11. Action Program for Rapid Washing Methods —J. M. JENSEN, *Michigan State College.*

D. Report of Quality and Marketing Committee—E. WALLENFELDT, *University of Wisconsin.*

E. Report of States with Exhibits and Committee on Teaching Ideas—R. T. HARRIS, *University of Wisconsin.*

F. Report of Type Rating Committee—J. W. LINN, *Kansas State College.*

G. Report of Dairy Cattle Health Committee—C. W. REEVES, *University of Tennessee.*

H. Report of Dairy Farm Management Committee—M. J. REGAN, *University of Missouri.*

11:00–12:00 **Joint Business Meeting**—Production and Extension Sections, *Room 206, H & F Bldg.*, E. C. SCHEIDENHELM, *Presiding.*

A. Report of Dairy Cattle Breeding Committee—E. J. PERRY, *Rutgers University.*

B. Report of Breeds Relations Committee—H. A. HERMAN, *University of Missouri.*

C. Report of Pasture and Roughage Committee—R. B. BECKER, *University of Florida.*

Afternoon Session

1:00– 4:00 **General Session**—Post War Problems of Dairying. (See General Program.)

4:00– 5:00 **Committee Meetings.**

Thursday, June 22

Morning Session

9:00–11:00 **Joint Symposium**—Production and Extension Sections, *Room 206, H & F Bldg.*

Mastitis and Feed Situation—DWIGHT ESPE, *Presiding.*

See General Program.

11:00–12:00 **Business Meeting**—Extension Section.

Afternoon Session

1:00– 3:30 **General Session**—Latin American Dairying. (See General Program.)

3:30 **General Business Meeting,** *Room 200, Campbell Hall.*

Evening

7:00 **Annual Banquet—Installation of Officers, Presentation of Borden Awards.**

PRODUCTION SECTION

Tuesday, June 20

Afternoon Sessions

DWIGHT ESPE—*Chairman*

1:00– 4:00 **Sectional Programs.**

SECTION A—Feeds and Feeding, *Room 100, B & Z Bldg.*, G. W. SALISBURY, *Presiding*

P1. Some Factors Affecting the Nutritive Value of Korean Lespedeza Hay—H. A. HERMAN, E. W. SWANSON, AND A. C. RAGSDALE, *University of Missouri.*

P2. The Feeding Value of Cheat for Dairy Cows—M. H. BERRY, K. L. TURK, AND L. A. MOORE, *University of Maryland.*

P3. Silage Fermentation Losses—A. E. PERKINS, *Ohio Agricultural Experiment Station.*

P4. Silage Versus Winter Pasture for Dairy Cows—C. E. WYLIE, B. P. HAZELWOOD, AND L. R. NEEL, *University of Tennessee.*

P5. Effect of Feeding Corn and Alfalfa Silage on the Fat- and Water-Soluble Vitamins—J. J. STEFANIAK, I. W. RUPEL, AND W. H. PETERSON, *University of Wisconsin.*

P6. New Feeding Standard for Milk Production—T. A. BAKER, *University of Delaware.*

P7. Limited Grain Ration Versus All Roughage Ration for Dairy Cattle—F. B. WOLBERG, A. A. SPIELMAN, V. L. MILLER, AND U. S. ASHWORTH, *State College of Washington.*

P8. Limited Grain Feeding of Dairy Cows—C. E. WYLIE, S. A. HINTON, AND B. P. HAZELWOOD, *University of Tennessee.*

P9. Carotene Losses in Freshly Cut Plant Tissues—R. K. WAUGH, S. M. HAUGE, AND J. H. HILTON, *Purdue University.*

P10. Vitamin A Requirements of Dairy Cattle for Normal Growth and Reproduction—J. H. HILTON, J. W. WILBUR, AND S. M. HAUGE, *Purdue University.*

P11. Carotene Levels for Growth and Reproduction in Dairy Bulls—I. R. JONES, *Oregon State College.*

P12. Changes in Blood-plasma Carotene Associated with Parturition and Lactation of Jersey Cows—A. H. KUHLMAN AND W. D. GALLUP, *Oklahoma A & M College.*

P13. Vitamin E in the Nutrition of Cattle—T. W. GULLICKSON, L. S. PALMER, W. L. BOYD, AND F. C. OLSON, *University of Minnesota.*

Paper to be Read by Title

P13a. The Value of Fat in Alfalfa Hay Rations—I. R. JONES, *Oregon State College.*

SECTION B—Physiology and Nutrition, *Room 209, B & Z Bldg.*, DWIGHT ESPE, *Presiding*

P14. Studies on Ketosis in Dairy Cattle—J. C. SHAW, *University of Connecticut.*

P15. Symptoms of Scurvy Observed in a Herd of Dairy Cattle—C. W. DUNCAN, C. F. HUFFMAN, R. MITCHELL, JR., AND J. T. REID, *Michigan State College.*

P16. The Relation of Vaccum to Speed in Mechanical Milking—VEARL SMITH AND W. E. PETERSEN, *University of Minnesota.*

P17. Studies of Mammary Gland Carbohydrate Metabolism in Vitro—C. B. KNODT AND W. E. PETERSEN, *University of Minnesota.*

P18. Pre-partum Milking—E. A. KEYES, J. J. REID, A. A. BORLAND, A. L. BEAM, AND P. S. WLLIAMS, *Pennsylvania State College.*

P19. The Use of Simplified Diets in the Study of the Fat Metabolism of the Mammary Gland—O. W. KAUFMANN, *University of Connecticut.*

P20. The Effect of Two Different Methods of Feeding Cod Liver Oil on Fat Test in Milk—L. A. MOORE, G. T. HOFFMAN, AND M. H. BERRY, *University of Maryland.*

P21. Further Observations on the Initiation and Maintenance of Lactation in Dairy Cattle—RALPH P. REECE, *New Jersey Agricultural Experiment Station.*

P22. The Response of Louisiana Milk Cows to Iodinated Casein Feeding—D. M. SEATH, CECIL BRANTON, AND A. H. GROTH, *Louisiana State University.*

P23. Studies in the Utilization of Thyroprotein by Ruminants—C. W. TURNER AND E. P. REINEKE, *University of Missouri.*

P24. Studies of Thyroid Physiology by Use of Thiourea and Its Derivatives—E. P. Reineke, A. B. Schultze, and C. W. Turner, *University of Missouri.*

P25. The Effect of Pitocin on Milk Lipase—Philip L. Kelly, *University of Arkansas.*

P26. Factors Affecting the Chylomicron Count—Dwight Espe and C. Y. Cannon, *Iowa State College.*

Paper to be Read by Title

P27. Morphology of the Teat in Relation to Milking and Trauma—W. E. Petersen, C. B. Knodt, and W. L. Boyd, *University of Minnesota.*

4:00– 5:00 **Committee Meetings.**

7:00 **Symposium**—Relation of Body Size to Milk Yield. (See General Program.)

Wednesday, June 21

Morning Session

9:00–11:00 **Sectional Programs.**

Section A—Feeds and Feeding—Calf Rearing—Harvesting Roughages, *Room 100, B & Z Bldg.*, Dwight Espe, *Presiding*

P28. Threshed Peanut Hay as a Roughage for Dairy Cows—A. H. Kuhlman and H. W. Cave, *Oklahoma A & M College.*

P29. The Use of Urea in Commercial Dairy Feeds—W. H. Hastings, *Nutritional Director, Lindsey-Robinson & Company.*

P30. Corn Silage Made with the Addition of Urea and Its Feeding Value—T. E. Woodward, *Bureau of Dairy Industry.*

P31. Urea Sorghum Silage—George K. Davis, C. L. Comar, R. B. Becker, and P. T. Dix Arnold, *University of Florida.*

P32. Urea Treated Corn Silage Versus Untreated Corn Silage as a Dairy Feed—G. H. Wise, J. H. Mitchell, J. P. LaMaster, and D. B. Roderick, *Clemson Agricultural College.*

P33. The Minimum Protein Requirement of Young Holstein Calves—L. E. Harris and J. K. Loosli, *Cornell University.*

P34. Studies on Carotene Conversion in the Young Calf—Norman S. Lundquist and Paul H. Phillips, *University of Wisconsin.*

P35. Observations on the Pathology of Dairy Calves on Low Vitamin A Diets—E. A. KEYES, S. I. BECHDEL, W. T. S. THORP, AND N. B. GUERRANT, *Pennsylvania State College.*

P36. The Effect of Sulfa Drugs and Immunity Development Upon Blood Plasma Vitamins A and C in the Young Bovine—NORMAN S. LUNDQUIST AND PAUL H. PHILLIPS, *University of Wisconsin.*

P37. The Use of Stored Colostrum to Replace Marketable Milk for Calf Feeding—N. N. ALLEN, *University of Vermont.*

P38. Experiences with the Forage Crop Harvester in Making Grass Silage—C. F. MONROE, *Ohio Agricultural Experiment Station.*

P39. Development of the Barn Hay-drying System—C. E. WYLIE AND JOHN A. SCHALLER, *University of Tennessee.*

SECTION B—Animal Breeding and Reproduction, *Room 209, B & Z Bldg.*, G. W. SALISBURY, *Presiding*

P40. Congenital Muscular Contracture and Ankylosis in Dairy Cattle—A. A. SPIELMAN, O. J. HILL, AND E. C. MCCULLOCK, *State College of Washington.*

P41. The Optimum Emphasis on Dam's Records when Proving Dairy Sires—JAY L. LUSH, *Iowa State College.*

P42. Tendency Toward Mastitis in Two Cow Families—J. M. MURPHY AND K. O. PFAU, *New Jersey Agricultural Experiment Station.*

P43. The Effect of the Oral Administration of Chloretone to Bulls on the Fertility of the Semen Produced—IRVINE ELLIOTT, *Cornell University.*

P44. Storage of Bovine Spermatozoa in Diluents Containing Certain Amino Acids—C. E. KNOOP, *Ohio Agricultural Experiment Station.*

P45. Recent Observations on the Preservation of Bull Semen—HENRY A. LARDY AND PAUL H. PHILLIPS, *University of Wisconsin.*

P46. Further Studies of the Effect of Dilution Rate on the Fertility of Bull Semen Used for Artificial Insemination—G. W. SALISBURY, IRVINE ELLIOTT, AND N. L. VANDEMARK, *Cornell University.*

P47. Conception Rate in Dairy Cattle by Artificial Insemination at Various Intervals Before and After

Ovulation—G.W. TRIMBERGER AND H. P. DAVIS, *University of Nebraska.*

P48. The Methylene Blue Reduction Test and Its Relation to Other Measures of Quality in Bull Semen—G. W. SALISBURY, ERNEST MERCIER, AND N. L. VANDEMARK, *Cornell University.*

P49. The Influence of Pregnancy on the Body Weight of Dairy Cows—D. N. PUTNAM AND H. O. HENDERSON, *University of West Virginia.*

P50. Factors to Consider in Long Distance Semen Shipping—H. A. HERMAN AND E. W. SWANSON, *University of Missouri.*

11:00–12:00 **Joint Business Meeting**—Production and Extension Sections, *Room 206, H & F Bldg.*, E. C. SCHEIDENHELM, *Presiding.*

A. Report of Dairy Cattle Breeding Committee—E. J. PERRY, *Rutgers University.*

B. Report of Breeds Relations Committee—H. A. HERMAN, *University of Missouri.*

C. Report of Pasture and Roughage Committee—R. B. BECKER, *University of Florida.*

Afternoon Session

1:00– 4:00 **General Session**—Post War Problems of Dairying. (See General Program.)

4:00– 5:00 **Committee Meetings.**

Thursday, June 22

Morning Session

9:00–11:00 **Joint Symposium**—Production and Extension Sections, *Room 206, H & F Bldg.*

Mastitis and Feed Situation—*See General Program.*

11:00–12:00 **Business Meeting**—Production Section, *Room 100, B & Z Bldg.*

Afternoon Session

1:00– 3:30 **General Session**—Latin American Dairying, *Room 200, Campbell Hall.*

3:30 **General Business Meeting,** *Room 200, Campbell Hall.*

Evening

7:00 **Annual Banquet—Installation of Officers and Presentation of Borden Awards.**

MANUFACTURING SECTION

Tuesday, June 20

Afternoon Session

P. F. SHARP—*Chairman*

1:00– 4:00 **Sectional Programs.**

SECTION A—Bacteriology, *Room 302, Campbell Hall*

M1. Factors Which Influence the Apparent Survival of Heat-Treated Bacteria—F. E. NELSON, *Iowa State College.*

M2. A Correlation of the Resazurin Test Read with an All-Purpose Lovibond Comparator at 10, 30, and 60 Minutes with the Standard Plate Count of Milk—S. IRENE JORGENSEN AND N. S. GOLDING, *State College of Washington.*

M3. Mastitis and the Plate Count of Milk—E. O. ANDERSON, *University of Connecticut.*

M4. Behavior of Bacteria in Certain Gases under Pressure—M. J. PRUCHA, *University of Illinois.*

M5. Bactericidal Property of Some Acids, Wetting Compounds, and Acid Cleaners—M. J. PRUCHA, *University of Illinois.*

M6. Farm Sources of *Oospora Lactis*—E. R. GARRISON, *University of Missouri.*

M7. Further Observations on the Use of the Propionates as Inhibitors of Mold on the Surface of Butter—J. C. OLSON AND H. MACY, *University of Minnesota.*

M8. The Mold Mycelia Count as an Index of Quality of Butter—LT. P. R. ELLIKER AND B. E. HORRALL, *Purdue University.*

M9. An Aerobacter Species in Whey Cream as a Cause of a Medicinal Flavor Encountered in Butter—T. J. CLAYDEN, *University of Arkansas.*

SECTION B—Chemistry, *Room 200, Campbell Hall*

M10. Some Chemical and Physical Properties of Washing Powder—P. S. LUCAS, *Michigan State College.*

M11. Factors Entering into the Testing of Some Cream for Extraneous Matter—K. M. RENNER, *Texas Technological College.*

M12. The Problem of Extraneous Matter in Cheddar Cheese—W. V. PRICE AND RAYMOND MIERSCH, *University of Wisconsin.*

M13. Correlating Lactic Acid Determination with Practical Milk Quality Test in Grading Milk for Manufactured Dairy Products—I. A. GOULD, *Michigan State College.*

M14. Use of Conductivity Measurements for Detecting Neutralization of Sweet Cream Buttermilk—S. T. COULTER AND R. W. KUNKEL, *University of Minnesota.*

M15. An Improved Technique for the Determination of the Volatile Acidity of Cheese—K. L. SMILEY AND A. C. DAHLBERG, *Cornell University.*

M16. A Study of Some of the Substances Adsorbed on the Fat Globules of Milk—ROBERT JENNESS, *University of Minnesota.*

M17. Some Factors Affecting the Inversion of Sucrose—T. R. FREEMAN AND E. L. FOUTS, *University of Florida.*

M18. Study of the Peroxide Value of Stored Spray-Dried Whole Milk Powder—HARRY PYENSON, P. H. TRACY, AND J. E. TRIMBLE, *University of Illinois.*

M19. The New Method for the Determination of Lipase in Milk. PHILIP L. KELLY, *University of Arkansas.*

4:00– 5:00 **Committee Meetings.**

Wednesday, June 21

Morning Session
P. F. SHARP—*Chairman*

9:00–11:00 **Sectional Programs.**

SECTION A—Butter and Cheese, *Room 203, Campbell Hall*

M20. The Influence of Butter Cultures and of Butter Flavors on the Quality of Butter—H. C. OLSON AND P. E. JOHNSON, *Oklahoma A and M College.*

M21. Effect of Various Bacteria on Diacetyl Content and Flavor of Butter—LT. P. R. ELLIKER, *Purdue University.*

M22. The Effect of Storage Temperatures on the Keeping Quality of Butter—C. W. STARK, J. R. CAMPBELL, AND E. S. GUTHRIE, *Cornell University.*

M23. Butter Studies—E. S. GUTHRIE, *Cornell University.*

M24. Action of Lipases from Various Sources on the Fat of Cheddar Cheese—F. J. BABEL, *Iowa State College.*

M25. Combining Lactic and Bulgaricus Fermentation in Cheese Making to Prevent Gas Formation in Cheese Ripening—N. S. GOLDING, *State College of Washington.*

M26. Does the Growth of Blue Mold in Roquefort Type Cheese Change Its Bacterial Flora?—N. S. GOLDING, *State College of Washington.*

M27. Effect of Heat Treatments of Milk on Quality and Ripening of Cheddar Cheese—A. O. CALL AND W. V. PRICE, *University of Wisconsin.*

M28. Relation of Corn and Alfalfa Silage to the Quality of Cheese and Its Carotene and Vitamin A Content—W. V. PRICE, K. HIGUCKI, AND W. H. PETERSON, *University of Wisconsin.*

SECTION B—Milk and Other Dairy Products,
Room 200, Campbell Hall

M29. Two Years' Experience in Deaerating Milk—E. S. GUTHRIE, *Cornell University.*

M30. The Relationship of the Individuality of the Cow to the Production of Rancid Milk—W. A. KRIENKE, *Oklahoma A & M College.*

M31. Milk as a Frozen Food—J. G. LEEDER AND F. J. DOAN, *Pennsylvania State College.*

M32. The Utilization of Skimmilk in Ice Cream Mix—W. S. ARBUCKLE, C. N. SHEPARDSON, AND H. M. WALLING, *A & M College of Texas.*

M33. Factors Affecting the Oxygen Content of the Gaseous Phase of Packaged Whole Milk Powder—JOHN HETRICK AND P. H. TRACY, *University of Illinois.*

M34. The Keeping Quality of Commercial Dried Whole Milks Packaged in Air and in Nitrogen—G. R. GREENBANK, P. A. WRIGHT, AND E. F. DEYSHER, *Bureau of Dairy Industry, U.S.D.A.*

M35. Further Observations Dealing with the Behavior of Ascorbic Acid in Evaporated Milk—D. V. JOSEPHSON AND F. J. DOAN, *Pennsylvania State College.*

M36. A Comparison of Different Types of Sweetening Agents in the Preservation of Condensed Milk—W. A. HOSKISSON AND P. H. TRACY, *University of Illinois.*

M37. Iron Content of Evaporated Milk in Relation to Greenish-Dark Discoloration in Mixtures of Coffee

and Evaporated Milk—W. C. COLE AND N. P. TARASSUK, *University of California.*

M38. Can We Hold Our Wartime Marketing Gains in Post-War Adjustments—C. G. MCBRIDE, *Ohio State University.*

11:00–12:00 **Business Meeting**—Manufacturing Section, *Room 200, Campbell Hall.*

Afternoon Session

1:00– 4:00 **General Session**—Post War Problems of Dairying. (See General Program.)

4:00– 5:00 **Committee Meetings.**

Thursday, June 22

Morning Session

P. F. SHARP—*Chairman*

9:00–11:00 **Symposium—Dehydrated Milk and Milk Products,** *Room 200, Campbell Hall.*

See General Program.

11:00–12:00 **Business Meeting**—Manufacturing Section, *Room 200, Campbell Hall.*

Afternoon Session

1:00– 3:30 **General Session**—Latin American Dairying. (See General Program.)

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VOLUME XXVII MAY, 1944 NUMBER 5

JOURNAL OF DAIRY SCIENCE

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ABSTRACTS OF LITERATURE

T. S. SUTTON, Editor
Columbus, Ohio

MILK AND MILK PRODUCTS

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CONTENTS

Bacteriology
Breeding
Butter
Cheese
Chemistry
Concentrated and dry milk; by-products
Diseases
Feeds and feeding
Food value of dairy products
Herd management
Ice cream
Milk
Miscellaneous
Physiology

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Espe, D. L.
Frazier, W. C.
Fuller, S. A.
Glick, D. P.
Goss, E. F.
Henderson, J. L.
Huffman, C. F.
Irvine, O. R.
Josephson, D. V.
Lucas, P. S.
Lush, J. L.
Marquardt, J. C.
Mueller, W. S.
Price, W. V.
Pyenson, Harry
Reece, Ralph P.
Roberts, Henry E.
Thomsen, L. C.
Trout, G. M.
Waugh, R. K.
Webb, B. H.
Weckel, K. G.
Weiser, H. H.
White, G. C.
Yale, M. W.

JOURNALS

American Butter Review
American Milk Review
American Journal of Diseases of Children
American Journal of Physiology
American Journal of Public Health
Archives of Pediatrics
Australian Journal of the Council for Scientific and Industrial Research

Biochemical Journal

Canadian Dairy and Ice Cream Journal
Canadian Journal of Public Health
Canadian Journal of Research
Certified Milk
Cornell Veterinarian

Dairy Industries
Dairy World

Endocrinology

Food in Canada
Food Industries
Food Manufacture
Food Research

Ice and Refrigeration
Ice Cream Field
Ice Cream Review
Ice Cream Trade Journal
Industrial and Engineering Chemistry

Journal of Agricultural Research
Journal of Agricultural Science
Journal of American Medical Association
Journal of American Veterinary Medical Association
Journal of Animal Science
Journal of Bacteriology
Journal of Biological Chemistry
Journal of Dairy Research
Journal of Dairy Science
Journal of Endocrinology
Journal of Experimental Medicine
Journal of General Physiology
Journal of Genetics
Journal of Heredity
Journal of Industrial and Engineering Chemistry
Journal of Infectious Diseases
Journal of Milk Technology
Journal of Nutrition
Journal of Pathology and Bacteriology
Journal of Physical Chemistry
Journal of Physiology
Journal of Veterinary Research

Lancet
Le Lait

Milk Dealer
Milk Industry
Milk Plant Monthly

National Butter and Cheese Journal
New Zealand Journal of Science and Technology

Oil and Soap

Pacific Dairy Review
Proceedings of Society of Experimental Biology and Medicine

Refrigerating Engineering

Scientific Agriculture
Southern Dairy Products Journal

SPECIAL PUBLICATIONS

Federal Dairying and Bacteriological Establishment, Liebefeld, Berne, Switzerland

International Association of Ice Cream Manufacturers
International Association of Milk Dealers

National Institute for Research in Dairying, Reading, England
New York Association of Dairy and Milk Inspectors

Prussian Dairy Research Institute, Kiel, Germany

State Agricultural Colleges and Experiment Stations

The Royal Technical College, Copenhagen, Denmark

United States Department of Agriculture

ABSTRACTS OF LITERATURE

BOOK REVIEW

157. Handbook for the Etiology, Diagnosis and Control of Infectious Bovine Mastitis. IVAL ARTHUR MERCHANT AND R. ALLEN PACKER, Iowa State College. Published by Burgess Publishing Co., Minneapolis, Minn. 66 pages. $1.25.

The authors of this handbook should be commended for their effort to gather scattered information regarding an important animal disease, sift out a great deal of cumbersome detail and concentrate the important factual matter between the covers of a small handbook.

This work will be a useful addition to the libraries of teachers, students and practitioners. Predisposing influences, bacteriology, diagnosis, control and treatment of the disease are the important subjects discussed. Sufficient detail is included to make the work a useful guide in procedures and techniques. T.S.S.

BACTERIOLOGY

158. Effect of Increase in Acidity on Antiseptic Efficiency. OTTO RAHN AND JEAN E. CONN, N. Y. State Col. of Agr., Cornell Univ., Ithaca, N. Y. Jour. Indus. and Engin. Chem., Indus. Ed., *36*, No. 2: 185. Feb., 1944.

Benzoic acid, salicylic acid, and sulfurous acid are nearly a hundred times as efficient antiseptics in strongly acid solutions as they are in neutral solutions. The toxic principle of benzoic and salicylic acids is the undissociated acid molecule. Growth of a wine yeast was completely suppressed when the concentration of undissociated benzoic acid reached 25 mg. per 100 ml. or when 4 mg. of undissociated salicylic acid was present. When sulfur dioxide dissociates in water the HSO_3^- ions inhibit the multiplication of *B. coli* but not of yeast. The rapid death of yeast is brought about by 7 to 8 mg. of undissociated H_2SO_3 per 100 ml.; *B. coli* can tolerate nearly ten times as much. B.H.W.

BUTTER

159. Sanitation in Buttermaking. WENDELL VINCENT. Amer. Butter Rev., *5*, No. 8: 238–241. 1943.

The inspector and plant operator are responsible for practices affecting sanitation within the plant as well as with the finished product. Sediment testing of cream is necessary and straining of cream by means of moving cloths rather than by in-line filters should be advised. Use of polluted water

and regard of pasteurization as a cover for poor sanitation are common offenses. Neglect in making repairs by soldering, improper care of stuffing boxes, poor cleansing and rinsing, and insanitary handling of butter scraps result in the production of lower score butter. Cream shipped by cream stations and independent buyers is of much lower quality than that shipped by direct shippers in the ratio of 2:1. While producers have been urged to increase quantity no one has ever urged that this be done at the expense of quality. P.S.L.

160. Butter Outlook and Its Relationship to Milk Solids for Ice Cream for 1944. T. G. STITTS, Chief, Dairy and Poultry Branch, F.D.A., Washington, D. C. Ice Cream Trade Jour., *40*, No. 1: 12. Jan., 1944.

A comprehensive picture is drawn by the author of the government's effort to apportion the available milk supply to production of the various dairy products required for civilian, military and lease-lend purposes. Ice cream manufacture is not likely to be further restricted inasmuch as the dairy industry will of necessity "lean heavily" on the ice cream industry to absorb milk solids when the food emergency is over. Butter is likely to be short for the duration but supplies should not become much scarcer than at present. If milk production does not "fall off" too much and if conservation order, F D O 79, works as it should, no coupon rationing of fluid milk will become necessary. A huge amount of cream has been moving into cold storage which tends to dislocate the dairy products program. Much of this should go into butter but it is doubtful if it was stored for that purpose. On the whole, the dairy products program seems to be working quite satisfactorily. F.J.D.

CHEESE

161. A Program for Maintaining Cheese Quality. G. H. WILSTER, Oreg. State Col., Corvallis. Natl. Butter and Cheese Jour., *35*, No. 2: 16. Feb., 1944.

The program recommends: grading milk by appearance, sediment test, flavor, methylene blue and curd tests; washing, sterilizing and drying cans; pasteurization of milk; the use of an active starter; limitation of acid development during manufacture to conform to a making period of about 5 hours from setting to milling; strict sanitation in factory maintenance; and careful curing at temperatures selected to bring out the best flavors. W.V.P.

162. Did You Ever See a Dream Working? PAUL MANDT, Olsen Pub. Co., Milwaukee, Wis. Natl. Butter and Cheese Jour., *35*, No. 2: 8. Feb., 1944.

A practical version of an ideal cheese factory is described and discussed. This well-equipped plant includes such unusual machinery as an autoclave

for starter cultures, a mechanically refrigerated starter can and glass holding tubes combined with a plate-type, regenerative pasteurizer. Cleanliness is emphasized in plant maintenance and is accomplished by systematic routines, simple instructions and assignment of responsibility to individuals. The plant operates with dry floors during the day. Making operations are run "by the clock" because of starter control methods and pasteurizing efficiency. W.V.P.

163. Some Causes of Slow Production of Acid During Cheesemaking. G. H. WILSTER, Oreg. State Col., Corvallis. Natl. Butter and Cheese Jour., *35*, No. 3: 10. March, 1944.

The common trouble of a starter "going dead" may be caused by bacteriophage, an unidentified agent which "eats" starter bacteria. Phage may be found in sewage, faeces, pus, and the dust and whey in a cheese factory. It gets into starter by contamination with mist from the whey separator, factory air and dust. The infection may last for months in a factory. Phage is destroyed by treatments which destroy the organisms which it attacks. Prevention of phage infection may be accomplished by disinfecting the factory by spraying, by protection of starter cultures from air-borne phage and by preparation of starter in a separate building designed to exclude phage-laden air. W.V.P.

164. A Program for Maintaining Cheese Quality. G. H. WILSTER, Oregon State Col., Corvallis, Oreg. Amer. Butter Rev., *5*, No. 9: 270–272, 283–284. 1943.

In view of the precarious situation in the cheese industry due to the manufacture in many cases of a low quality of product the author has outlined and discussed rather completely nine factors important in the improvement of cheese. These are: 1, the grading of milk and rejection of poor quality product; 2, thorough cleansing and proper care of cans after washing; 3, efficient pasteurization of milk; 4, proper use of a good starter; 5, unhurried methods of manufacture with especial attention given to acid development; 6, sanitation; 7, curing at low temperatures and moderate humidity; 8, regulation of moisture and fat content through adequate and regular laboratory control; and, 9, grading as a means of improvement. P.S.L.

CHEMISTRY

165. The Total Nitrogen Content of Egg Albumin and Other Proteins. A. C. CHIBNALL, M. W. REES, AND E. F. WILLIAMS, Biochem. Dept., Imperial Col., London, S.W. 7. Biochem. Jour., *37*, No. 3: 354–359. Sept., 1943.

With the development of new catalysts for the Kjeldahl nitrogen determination, and especially with the advent of the micro Kjeldahl method, there

has been a tendency to reduce the time required for digesting the sample. The authors feel that this has been done at the expense of accuracy, for the rapid digestion gives lower percentages of nitrogen than those reported by such earlier workers as Osborne and Campbell, as well as their own results with longer digestion periods. "With proteins and protein hydrolysates it is necessary to continue the heating for 8 hours or more after the digest has cleared." In reporting nitrogen determinations on proteins there should be more details of the procedure given, as well as some history of the treatment of the protein or its method of preparation. This may help to intelligently interpret the results.

For casein, prepared by the method of Cohn and Hendry they report 15.73% nitrogen on a moisture-free and ash-free basis. For B-lactoglobulin, which was twice recrystallized, coagulated by heat and washed free from inorganic salts and dried, they report a value of 15.58% nitrogen on a moisture-free and ash-free basis.

Abstractor's note: The term B-lactoglobulin is not frequently used in literature in America. There is a need for a unified system of nomenclature in the field of the milk albumins and globulins. The B-lactoglobulin referred to above is apparently that fraction of the milk albumin which was crystallized by A. D. Palmer. This is the only milk protein reported to have been crystallized to date. A.O.C.

166. The Dicarboxylic and Basic Amino Acids of Edestin, Egg Albumin and B-Lactoglobulin. A. C. CHIBNALL, M. W. REES, AND E. F. WILLIAMS, Biochem. Dept., Imperial Col., London, S.W. 7. Biochem. Jour., *37*, No. 3: 372–388. Sept., 1943.

A detailed procedure for the estimation of dicarboxylic and basic amino acids is given. The method is admittedly a long one, requiring about 900 working hours, but the authors state that the results are more reliable than those of any method reported heretofore, accounting for all but 1.25% of the total protein nitrogen. The method differs from those usually used in that no reagent is added to the hydrolysate unless it can be quantitatively removed later without carrying with it an appreciable amount of protein.

For B-lactoglobulin the percentages of the total nitrogen as the dicarboxylic acids glutamic and aspartic are given as 13.14% and 6.68% respectively, while the basic amino acids are: arginine 5.95% histidine 2.69% and lysine 12.07% of the total nitrogen. A.O.C.

167. Oxidative Rancidity in Edible Fats. L. R. BRYANT, Ont. Agr. Col., Guelph, Ontario. Food in Canada, *4*, No. 1: 7. 1944.

Atmospheric oxygen produces a type of rancidity in food fats characterized by changes in color, destruction of the fat-soluble vitamins and the development of off-flavors. The chemical make up determines the suscepti-

bility of various fats to this chemical change. Aside from the fat itself, such factors as temperature, light, ozone, metals or the presence of anti-oxidants accelerate or inhibit the reaction rate. Tests for the susceptibility to, and degree of, oxidation are described. O.R.I.

168. The Causes, Cures and Methods of Preventing Rancidity. C. H. CASTELL, Ont. Agr. Col., Guelph, Ontario. Food in Canada, *3*, No. 10: 11; *3*, No. 11: 11; *3*, No. 12: 10. 1943.

This series of three articles deals with fat spoilage from the standpoint of (1) hydrolytic rancidity of non-microbial origin, (2) rancidity and off-flavors produced by yeasts, molds and bacteria of the aerobic group, and (3) off-flavors of a similar character produced by the butyric-acid-forming anaerobes. In the first of these, the chemical make-up of fats is described with stress being placed on strong odors and flavors possessed by some of the free fatty acids. The sources and characteristics of the lipases are discussed and methods of measuring rancidity and the lipase content of foods described. Particular attention is given to milk and dairy products.

In the second article dealing with microbial agencies causing food spoilage it is pointed out that oxidative rancidity is often an important secondary reaction occurring after the fat has first been attacked by bacteria. Yeasts are very rarely lipolytic but most molds produce lipase. Fat-splitting bacteria are numerous and widely distributed and many species grow at low temperatures. Members of the *Alcaligines, Aerobacter, Achromobacter, Pseudomonas* and *Serratia* genera are of greatest importance. Some of the newer methods whereby organisms of this group may be identified or counted are described.

Butyric-acid-forming anaerobes produce a type of rancidity in some foods entirely unrelated to the fat content. These organisms are strict anaerobes, and produce heat-resistant spores. Butyric acid is one of the chief products produced when carbohydrates are fermented. Relatively few species grow in an acid medium or at low temperatures. Culturing and counting require the use of either an anaerobic jar or growth in a corn-liver or cereal grass medium. Dairy products, particularly some European varieties of cheese, sometimes develop rancid flavors as a result of the growth of organisms of this group. O.R.I.

CONCENTRATED AND DRY MILK; BY-PRODUCTS

169. Improving Keeping Quality of Dry Whole Milk. C. D. DAHLE AND D. V. JOSEPHSON, Dairy Dept., Penn. State Col. Milk Plant Monthly, *32*, No. 10: 28–29. 1943.

By removal of a large percentage of lecithin through churning and separating out the butter oil and through supercentrifuging the skimmilk,

atmospheric roll dry whole milk of good keeping quality could be made from the recombined product. The possibility of removing some other prooxidant than lecithin by the supercentrifuging treatment was suggested by the great improvement in keeping quality noted. The prevalent flavor defect occurring in dry whole milk is of an oxidative type. G.M.T.

FOOD VALUE OF DAIRY PRODUCTS

170. Determination of Vitamin A and Carotene in Milk. A Rapid Extraction Procedure. PAUL D. BOYER, ROBERT SPITZER, CURTIS JENSEN, AND PAUL H. PHILLIPS, College of Agr., Univ. of Wis., Madison, Wis. Jour. Indus. and Engin. Chem., Analyt. Ed., *16*, No. 2: 101. Feb., 1944.

The authors describe a rapid procedure for the extraction and determination of vitamin A and carotene in milk. Two volumes of milk mixed with 3 volumes of alcoholic potassium hydroxide are allowed to stand for 3 hours at room temperature. The mixture is then extracted twice with ether and the vitamin A and carotene are determined by means of the Carr-Price reaction and with the aid of an Evelyn photoelectric colorimeter. The new procedure was compared to the longer procedure of Olsen, Hegsted, and Peterson. The comparative analyses showed that with a single ether extraction the new method occasionally gave low results for vitamin A. The double extraction procedure which was described gave good checks with both pasteurized and raw milks from cows of various breeds. The carotene determination as given in the procedure is a measure of the total carotenoids in the milk. B.H.W.

171. Determination of Vitamin A and Carotenoids in Butterfat. Comparison of Direct Spectrophotometry with Filter Photometry and Use of the Antimony Trichloride Reaction. F. P. ZCHEILE, H. A. NASH, R. L. HENRY, AND L. F. GREEN, Purdue Univ. Agr. Expt. Sta., Lafayette, Ind. Jour. Indus. and Engin. Chem., Analyt. Ed., *16*, No. 2: 83. Feb., 1944.

The data reported were obtained during a comparative study of methods for the determination of vitamin A and carotenoids in butter by the Technical Committee on Vitamin A Researches in cooperation with the National Cooperative Project on the Vitamin A Potency of Market Butters. Six representative samples of butterfat from sweet cream were prepared and analyzed by seven collaborators. This paper compares the results of the direct spectroscopic method used by the authors and the colorimetry methods used by the other collaborators. Comparison of the carotenoid content of the butterfats determined by the different methods showed that from the total of 42 determinations, only 5 deviated from their corresponding means

by more than 7%, the maximum deviation being 13.3%. The direct spectrophotometry results had both the smallest mean absolute deviation and the smallest maximum deviation. Comparison of the determinations of vitamin A contents showed the results from direct spectrophotometry to have mean absolute and maximum deviations two-thirds as great as the over-all averages. The antimony trichloride reaction was considered the preferred physico-chemical method available for butters containing azo dyes.

B.H.W.

172. Some Experiments on the Possible Relationship between Vitamin C and Calcification. GEOFFREY H. BOURNE, Univ. Lab. of Physiol., Oxford. Jour. Physiol., *102*, No. 3: 319. Dec., 1943.

"Bone salt does not appear to be deposited (except in severe scurvy) until there is an adequate fibrous matrix to receive it. The production of the fibrous matrix of bone and the deposition of bone salt are therefore simultaneous processes. It would seem that as long as there is sufficient vitamin C to produce matrix then that matrix will be calcified. The function of vitamin C in bone formation appears to be to facilitate the production, not just of bone matrix, but of bone matrix impregnated with phosphatase. There is no evidence that vitamin C can be regarded as a coenzyme of phosphatase in calcificatory processes. The apparent reduction of phosphatase activity in scurvy is therefore probably due to a reduction in the amount of bone matrix produced.

"That it is actually vitamin C and not some associated impurity that is responsible for this is suggested by the fact that the administration of vitamin P (citrin) and sodium citrate did not result in the formation of more osteoid trabeculae or the deposition of more bone salt than vitamin C alone."

D.E.

HERD MANAGEMENT

173. Preparing the Cow for Milking. W. E. PETERSEN, Prof. of Dairy Husbandry, Univ. of Minn. Milk Plant Monthly, *32*, No. 12: 26–27. 1943.

"Letting down" of milk is the result of a positive act causing tiny muscle cells to contract thus squeezing the milk out of the alveoli where it is made. This is a reflex act, spontaneous to a stimulus, which causes the pituitary gland to secrete a hormone (oxytocin) into the blood by which it is carried to the mammary gland.

The following fundamentals about the response and action of this reflex furnish the basis for several recommended practices: 1. Response to the stimulus is interfered with by any condition that distracts the cow. 2. Once the "let down" has occurred, it is effective for but a short period of time.

3. Cows may become conditioned to let down their milk to a number of different stimuli. 4. The way a cow responds to milking is determined largely by training. 5. About 45 seconds are required from the application of the stimulus to the "let down" of the milk.

The following 11 rules are based on these fundamentals: 1. Handle heifers carefully when they are first milked. 2. Avoid the unusual during milking. 3. Do not treat the cow roughly at any time. 4. The milker must be a person who does not arouse the cow's suspicion. 5. Do not wash or massage the udders or stimulate cows in other ways to let down their milk before milking is to begin. 6. Milk those cows first which let down their milk in response to preparations for milking. 7. Milk rapidly. 8. Do not practice prolonged stripping. 9. Operate milking machines according to the manufacturers' directions. 10. Do not leave the milking machine on the cow after the milk has ceased flowing. 11. Develop the technique of knowing by feel when the gland has been emptied of milk.

For the best operation of milking machine the following 4 points must be observed: 1. Stimulate the cow to let down her milk about one minute before attaching the machine. 2. Operate the machine according to the manufacturer's directions. 3. As soon as the teat cups begin crawling upward begin stripping by tugging downward with sufficient force to lower the teat cups part way down on the teat and at the same time stroke each quarter downward in succession with the free hand. 4. Remove the machine as soon as milk ceases flowing. G.M.T.

ICE CREAM

174. Ice Cream with the A.E.F. S/Sgt. J. A. Rabuffo, E.T.O., England. Ice Cream Trade Jour., *40*, No. 2: 24. Feb., 1944.

After mentioning the shipment of ice cream freezers, fountains, ice cream mix and accessories which have been authorized for distribution and installation in the European Theater of Operations to supply Uncle Sam's soldiers with fountain products which they have greatly missed, "G. I. Joe" tells of the ingenuity of the cooks and "K.P.'s" in improvising ice cream freezers and ice cream mixes under all manner of circumstances. He admits that results rarely compare favorably with the ice cream to which they have been accustomed but are far better than none. In many cases arrangements have been made with local ice cream makers (who no longer make) to use freezing equipment, if the commissaries are able to provide some form of mix ingredients. "G. I. Joe" presents one cook's closely guarded secret formula, revealed in a weak moment, as being composed of: evaporated milk, powdered milk, water, sugar, powdered eggs, corn starch and a little salt. The concoction is brought to a boil, cooled and frozen. F.J.D.

175. Pooling Cabinet Service. W. H. SNEATH, William Neilson, Ltd., Toronto, Canada. Ice Cream Trade Jour., *40*, No. 2: 30. Feb., 1944.

A pooled service for checking, adjusting and repairing refrigerated cabinets belonging to Canadian ice cream manufacturers is explained. The system was applied throughout the country by establishing zones, and all prices for parts, mileage rates, hourly wages, etc., were made uniform for each zone. One company was made responsible for all services in each zone and while the yearly records have not been compiled and summarized, there is little question but what tremendous savings have been achieved in gasoline, tires, mobile equipment, manpower and in the avoidance of breakdown. It is emphasized that successful operation of such a cooperative service can only be obtained through the confidence and cooperation of the entire personnel as well as the organizations involved. F.J.D.

176. Sherbets—What About Their Future? V. M. RABUFFO, Ice Cream Trade Jour., New York City. Ice Cream Trade Jour., *40*, No. 2: 16. Feb., 1944.

With an estimated production of 75,000,000 gallons of sherbets and ices forecast for 1944, the author reviews the experiences of the ice cream industry during 1943, pointing out the errors made in "forcing" these products on dealers as a result of FDO–8 rather than preparing the way by a "selling" campaign. Opinions and experiences are cited and the potentialities of sherbets and ices in the post war picture are stressed. It is pointed out that much progress has lately been made in improving the quality of sherbets and ices and since there is a strong possibility of continued rationing of milk solids, for a period of time after the war, it is suggested that the industry should attempt to correct its recent error by engaging in an extensive effort to sell consumers on the "goodness" and palatability of these milk-solids-sparing products. Sherbets and ices play too vital a role and represent too large a volume to be neglected. The situation is a challenge to the ice cream industry. F.J.D.

177. The Bacterial Content of Shell Eggs. C. K. JOHNS, Bact. and Dairy Research, Dom. Dept. of Agr., Ottawa, Canada. Food in Canada, *3*, No. 12: 15. 1943.

Bacteria count limits were incorporated into the 1943 Canada–Great Britain dried egg contracts. To learn whether or not shell eggs contribute materially to the count of the powder, bacterial numbers were determined on whole eggs by smearing 1/2000 g. loopfuls of mixed egg on tryptone glucose extract milk agar slants. Ninety per cent of the 348 eggs examined gave counts under 10,000 per g. with only one egg in excess of 500,000 per g The eggs were all 2 months old when examined. O.R.I.

178. Concrete in Ice Cream Plants. LAWRENCE FLYNN. Ice Cream Field, *43*, No. 1. Jan., 1944.

Floors which give the best service in food plants must have a top surface which wears well and resists the attacks of fruit juices, sugar solutions, and lactic acid, the author states.

Floors with porous surfaces are much more readily affected by acids and sugar solutions than are those with impervious surfaces. Special treatments can be employed to make porous surfaces practically impervious.

Dairy and ice cream plant floors are subject to hard usage and should be heavy duty concrete floors. The following basic principles are listed as essential in producing wear-resistant floor finishes: 1. Use only suitable, clean materials. 2. Use not in excess of 4½–5 gallons of mixing water per sack of cement. 3. Avoid "segregation" resulting in free water and fine material on top surface. 4. Keep concrete damp as long as possible. At least a week is required for normal Portland cement and three days when high early Portland cement is employed.

The importance of selecting suitable aggregate material and the use of proper proportions of ingredients as well as employing the proper methods and procedures are stressed. To avoid cracking of floor surfaces the author suggests the use of light wire mesh, 4 × 4 inch, No. 10 gauge wire weighing 31 pounds per 100 sq. ft. near the middle of the top or wearing course, and to avoid severe wear it is considered important to have as much coarse aggregate as possible near the surface.

Two methods are described for treating floor surfaces in order to make them impervious. One treatment consists in the application of warm linseed oil, Chinawood oil, or soy bean oil. To assist penetration the first coat should be thin (equal parts of oil and turpentine or other thinner). A second application can be given with a greater proportion of oil to thinner after the first coat has dried. A second method is the application of paraffin. It should have a melting point of 150° F. and should be applied as a paste prepared by melting 4 parts paraffin with 1 part turpentine and 16 parts toluol. Apply with brush and allow to penetrate 24 hours, keeping the floor warm; then polish with polishing machine.

Directions are also given for re-surfacing old floors after chipping away the old concrete to a depth of 1 inch. W.C.C.

MILK

179. Influence of Temperature in Open and Closed Truck Hauling. C. M. PESCK, Dairy and Food Dept., Minneapolis, Minn. Amer. Butter Rev., *5*, No. 9: 274–276. 1944.

Roof temperature under the same conditions was 16° lower on a cab made from aluminum-painted wood than on black iron. During each stop

roof temperature increased 6°. Cans of cold water and cream appreciably affected the temperature in the truck, lowering it to 70° when the outside temperature at the start was 92°, and 97° at the end of the trip.

With an open truck and cans covered with a canvas the temperature on the floor of the truck was 85° when the cans were first loaded and 88° on arrival at the creamery. Outside temperature was 86° at the start of the trip, 94° at the end. Temperature varied with the wind direction and did not steadily decrease as in the closed truck. Acidity of the cream sharply increased in the canvas-covered open truck. In the original article an accompanying table gives detailed results. P.S.L.

180. Testing Homogenized Milk. S. T. COULTER, Univ. of Minn., Minneapolis, Minn. Amer. Milk Rev., *5*, No. 12: 382. 1943.

The method previously proposed by J. C. Marquardt of the New York (Geneva) Agricultural Experiment Station was given several trials at the Minnesota Dairy Department and gave fairly satisfactory results. The method developed is one of modifying the ordinary Babcock procedure. P.S.L.

181. Bad Flavors in Milk. E. S. GUTHRIE, Cornell Univ., Ithaca, N. Y. Amer. Milk Rev., *5*, Nos. 11 and 12: 338–346, and 372–385. 1943.

This article combines in an interesting and helpful manner the experimental work of many writers as regards flavors that may occur in milk, together with a discussion of conditions favoring their presence and controlling the degree to which they are present. Those discussed more fully are feed, rancid, lipolytic, those due to fat content and to treatment of milk, as homogenization, oxidized, metallic, cooked, disinfectant, and absorbed flavors. P.S.L.

182. Milk—Dairy Products Problems. LELAND SPENCER, Cornell Univ., Ithaca, N. Y. Amer. Milk Rev., *5*, No. 9: 276–282. 1943.

More milk has been produced yearly than on the average for 1935–9 but demand has been much greater than production. The military has taken 19% of the production ordinarily available. As compared with 1935–9, consumption by civilians was lower in 1942 by 7% but up 10% for fluid milk, 13% for canned milk, 14% for cheese, and 67% for ice cream. Besides these demands there are those of the export market. Price ceilings and shortage of labor and feed have added to production difficulties. Price regulations have caused in many cases price maladjustments that become disruptive. To cushion to as great extent as possible the effects of the probable shortage this fall the author suggests six remedies: 1, adjustment of price ceilings in markets operating at a disadvantage; 2, confine shipment of milk

within 500 mile distances; 3, grant priority to milk within its own milk shed; 4, ban all sales of cream if necessary; 5, limit fluid milk sales if necessary; 6, ration milk but only if absolutely necessary and as a last resort. P.S.L.

183. A Portable Resazurin Outfit. J. G. DAVIS, Natl. Inst. for Res. in Dairying, Shinfield, Reading. The Milk Indus., *24*, No. 6: 47. Dec., 1943.

A portable test kit has been developed for the application of the resazurin test in the field. The outfit is essentially designed for use on the farm and in small dairies. A picture of the apparatus and carrying case is shown and list of items contained in the apparatus is given.

It is advantageous to use the portable resazurin outfit directly after milking as positive results on freshly-drawn milk indicate the presence of body cells or a mastitis condition. The more severe the mastitis the quicker is the dye reduced or changed from blue to pink. It is also advantageous to run the resazurin test on the mixed herd milk as it comes into the plant, for resazurin reduction in such milk would probably be due to bacteria.

For quick resazurin tests on the farm the sample can be milked directly into the test tubes after removing the foremilk. It is recommended that individual quarters be tested. The rennet test is recommended for use in conjunction with the resazurin test. The methods for performing these tests are outlined.

Very bad samples will reduce resazurin rapidly and fail to clot with rennet. If a sample clots slowly with rennet but does not reduce resazurin, it may indicate a past history of mastitis. A change of color of resazurin without slow clotting may indicate an incipient infection which should receive treatment at once.

The following advantages of testing cows for abnormal milk are listed: a. Economic advantages. Infected cows are inefficient converters of food to milk. b. Danger of low solids-not-fat. The writer claims that the resazurin-rennet test can be correlated closely with solids-not-fat content. c. Danger of infecting other cows. The resazurin test may pick out bad cases of infection which may be segregated or disposed of or milked last. d. Effect on bacterial count. Abnormal milk usually increases bacterial count of mixed milk from herd. e. Effect of abnormal milk on manufacture of milk products. Abnormal milk produces weak body and slow starter in cheese manufacture.

The test can be used to good advantage in checking animals that are being purchased for addition to the herd. H.P.

184. Interpreting Bacterial Counts to Producers. O. A. GHIGGOILE, Chief, Bur. of Dairy Serv., Calif. State Dept. of Agr. Milk Plant Monthly, *32*, No. 10: 26–27. 1943.

The responsibility of producing good milk rests with the producer, who

should be instructed in a non-technical language about bacteria, their mode of entry into milk, their rate of multiplication, and the cleanliness and care of dairy equipment. The manner in which the producer is approached will do much in bringing about cooperation in producing a better grade milk. Once the producer realizes and accepts his full responsibility in producing high quality milk with a low bacterial count, he will be proud of his accomplishment. G.M.T.

185. How to Prevent and Remove Milk Deposits. LEWIS SHERE, Pres., The Diversey Corp. Milk Plant Monthly, *32*, No. 11: 32, 34, 39, 40. 1943.

Milk stone is a complex homogeneous mixture of organic and inorganic substances which adhere tenaciously to dairy equipment. It is unsightly, may cause off-flavors, interferes with heat transfer and is a source of high bacteria counts. The amount and composition of milk stone is influenced by: 1, the speed with which the milk flows through the equipment; 2, the final temperature to which the milk is heated; 3, the amount of milk handled; 4, type of dairy product handled; 5, type of equipment used; 6, hardness of the water; 7, nature of cleaners and sterilizers; and 8, time of rinsing.

Milk stone formation may be minimized by: 1. Rinsing the equipment with cold water; 2. Cleaning equipment thoroughly every day; 3. Rinsing the cleaning solution from the equipment; 4. Using chlorine sterilizers rather than heat; 5. Preventing corrosion of equipment.

Accumulated milk stone may be removed effectively and efficiently by using an acid-type product, following the manufacturer's directions. Precautions should be taken that the equipment is not damaged. G.M.T.

186. Interpreting Bacterial Counts to Producers. WALTER D. TIEDEMAN, Chief of Bur. of Milk Sanitation, N. Y. State Dept. of Health. Milk Plant Monthly, *32*, No. 10: 25–26. 1943.

Bacterial examination of milk is done primarily to improve the quality of the raw milk supply. Interpretations of the bacteria count should be made in a language the producers understand by one who appreciates the limitations of the laboratory tests. Since there is no advantage in reporting figures, the reporting of the following classes of bacteria is suggested: class 1—200,000 or less; class 2—from 200,000 to 1,000,000; class 3—over 1,000,000.

Milk falling in class 1 is satisfactory; that in class 2 needs improvement; while that in class 3 calls for immediate and definite action.

A similar classification of cell counts is suggested as follows: cells + = 500,000 or less per ml.; cells ++ = from 500,000 to 5,000,000; cells +++ = over 5,000,000.

A two-plus cell count might be interpreted as indicating the presence of mastitis in the herd, while a three-plus cell count is more definitely an indication of mastitis particularly when accompanied by long chain streptococci. The technician should help the dairyman by observing the type of bacteria and suggesting their significance. Repeating counts after unsatisfactory reports have been reported to see whether the report has been translated into action is good practice. G.M.T.

PHYSIOLOGY

187. The Blood Volume of Normal Animals. F. C. COURTICE. Jour. Physiol., *102*, No. 3: 290. Dec., 1943.

The blood volume of 30 goats has been estimated with the blue dye T–1824. The mean plasma volume was 53 cc. per kg. body weight and blood volume was 70 cc. per kg. The blood volume of goats as well as that of rabbits, dogs and horses is proportional to the body weight and not to surface area. The effect on blood volume of stage of lactation or productivity is not mentioned. The author does state that the blood volume of four highly trained greyhounds was much higher due to a higher cell volume. The blood volume depends upon the bulk of the animal tissue, especially muscle, and not upon the rate of metabolism. D.E.

188. Lipolysis and Fat Absorption. A. C. FRAZER, Physiol. Dept., St. Mary's Hosp. Med. School, London, and the Pharmacol. Dept., Univ. of London. Jour. Physiol., *102*, No. 3: 329. Dec., 1943.

The ingestion of neutral fat normally leads to a characteristic appearance of the intestinal cells, to milkiness of the lacteals, to a marked systemic lipemia and to deposition of fat in the fat depots. The addition of potent lipase to the ingested neutral fat causes small instead of large granules to appear in the intestinal cells, the lacteals remain almost clear, the systemic blood shows but a slight lipemia, and the deposition of the fats is much decreased. The portal blood and liver, which show only slight changes after neutral fat ingestion, exhibit marked lipemia and deposition respectively if lipase is added to the neutral fat. The results following the ingestion of neutral fat and lipase are thus similar to those seen after the administration of fatty acid. It is possible to suppress almost completely the post-absorptive systemic lipemia by the addition of lipase to the standard fat-containing meal. Lipolysis should be regarded as a determining factor in the fate of absorbed fat and possibly as a means of providing essential raw materials for the synthesis of lecithin and the formation of soaps.

The complete inhibition of lipolysis by a long chain sulphate, sodium cetyl sulphate, in rats, does not prevent triglyceride absorption. D.E.

189. Differentiation in the Absorption of Olive Oil and Oleic Acid in the Rat. A. C. Frazer, Physiol. Dept., St. Mary's Hosp. Med. School, London, and the Pharmacol. Dept., Univ. of Birmingham. Jour. Physiol., *102*, No. 3: 306. Dec., 1943.

According to the author's view, lipolysis is only partial in the intestinal tract of the adult rat, and hydrolysis of the triglyceride molecule is not regarded as an essential preliminary to its absorption. Fatty acid passes by the portal vein to the liver, while neutral fat goes by the lymphatic route to the systemic blood and thence to the main fat depots to be stored for future use. Stained fatty acids fed over a period of 10 days result in no staining of these areas, but rather, appear in the liver. The degree of lipolysis is, thus, a determining factor in the immediate fate of absorbed fat. D.E.

The NINTH *of a series discussing how Nature and Man together develop our finer foods.*

THE HIGHER MATHEMATICS OF *Flavoring*

"Season to taste"—that good old reliable cook-book recommendation was packed with good sense. But the modern food processor is a housewife whose dining room may extend for hundreds of miles from his kitchen. It takes more than fancy labels and simple rules to help him deliver a uniformly good product to his clamoring and hungry family of consumers.

He must have a realization, and a knowledge, of all the negatives as well as positives that affect flavor These higher mathematics of proper flavoring are perfectly illustrated in the case of vanilla ice cream.

Apparent loss of vanilla flavor, beginning a short time after the ice cream was made, had been somewhat of a mystery until recent years. Frequently it was referred to as "freezing out." Let us examine some of the clues, uncovered by leading authorities, that indicate how this disappearing act is performed.

The factors that must be taken into consideration are numerous. They start as far back as the feed the dairy cow consumes. Mere mention of the occasional garlicky taste in milk makes even laymen realize that feeds affect flavor in varying degrees.

Next, the effect of processing on flavor must be kept in mind. A cooked flavor may result from pasteurization, especially in localities whose regulations require high temperatures. Differences in the metals that make up the various processing units encourage electrolytic reactions. Homogenization spills the fat from out its protective membrane and may predispose this important material to the ravages of oxidation.

All these things are important because milk fats take up outside aromatics very readily. Every woman is familiar with this fact. She knows that exposed butter and milk desserts will take up the odors of many things in the refrigerator.

But the milk fats in ice cream also play tricks with added flavoring. They absorb more vanilla flavoring, for instance, than they will give out at the time the ice cream is eaten. Nothing drives this home so well as the fact that ice cream with 18% fat content requires almost half again as much flavoring material as ice cream with 12% fat content, to achieve the same apparent height of flavor.

All of these are simple addition and subtraction compared with the complex multiplication of problems imposed by:

—storage
—exposure at point of sale
—low temperature consumption.

In order to meet the public's demand, at a moment's notice, the ice cream industry can proudly claim one of the most comprehensive and efficient storage systems in the food business. But here, again, proper flavoring must anticipate the cumulative effect of all factors on flavor during storage and merchandising. A treatise could be written on this subject alone. One of the obvious points is that off-flavors and latent off-flavors do develop in quality dairy products, due to the milk itself and to the necessary processing, tending to build up and to overshadow the original vanilla flavoring, which does not actually disappear or "freeze out."

Therefore, students of the subject realize that the vanilla bean alone does not contain a sufficiently well-balanced quota of aromatics to overcome these unwanted flavors, and still be able to impart a full, well rounded vanilla flavor at the time the ice cream is eaten. This is one of a series of factors that has made it a trade custom to use added vanillin in the development of a truly fine and uniformly flavored vanilla ice cream.

No wonder the production of vanilla flavor is an art in itself. The knowledge that vanillin must be added to the vanilla bean is just the start. *How* to do it in the processing is another fine art. In flavoring, ***it's not only what goes into the mix, it is what comes out in the taste that counts!***

Published in the interest of continued progressive thinking in the food industries, by

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makers of Mixrum and other powdered vanilla products

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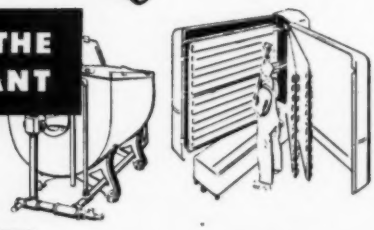
HELPS CONTROL ROPY MILK

ON THE FARM

IN THE PLANT



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DIVERSOL

Control and prevention of ropy milk begins on the dairy farm . . . likewise demands rigid measures of sanitation throughout the dairy plant. Both on the farm and in the dairy plant, Diversol, the quick-acting chlorine bactericide has proven most effective for all disinfecting operations.

Diversol comes in stable crystals that lock in the available chlorine until dissolved in water . . . thus assuring solutions of definite germicidal strength. Furthermore, Diversol's unusual penetrating power makes possible contact with bacteria. Harmless to all metal surfaces commonly used in dairy equipment . . . easy to use . . . leaves no film or scale. Write for technical bulletin "The Control and Prevention of Ropy Milk," **THE DIVERSEY CORPORATION, 53 W. Jackson Blvd., Chicago 4.**

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of definitely better flavor & aroma-producing qualities.

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Cheese Bandages, Circles
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MILWAUKEE, WISCONSIN

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In 1894 The Holstein-Friesian Association of America adopted its system of official records under supervision of State Experiment Stations and recognized and employed the newly invented Babcock Test for determining percentage of butterfat. Since the establishment of Advanced Registry more than 69,000 records have been accepted and recorded by The Holstein-Friesian Association of America.

In 1928 the program of HERD TEST was established, in which every cow in the herd was tested for production. Since that time more than 118,000 records have been completed in the Herd Test program to date. Also, 2,113 herd averages above 400 pounds of butterfat have been completed since that time.

As these tests have progressed attention has been focused on the significance of lifetime production. In the Herd Test and Advanced Register a total of 803 cows have credits above 100,000 pounds of milk, and of these 13 have produced more than 200,000 pounds.

Starting with the private record of "Dowager" in 1872 the Holstein breed has always maintained its leadership in the important field of production.

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For best results in the cheese vat, specify Marschall's. High in strength, pure and uniform.

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SUCCESSFUL Way to Keep Bacteria Counts Low!

"Bacteriological cleanliness" of equipment depends mainly on two IMPORTANT factors, (1) thorough removal of milk film and fats followed by (2) germicidal treatment. To meet these requirements more easily and effectively, first use specially formulated Oakite Dairy Detergents for cleaning processing equipment. THEN, for subsequent germicidal treatment use rapid-acting, POTENT

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This material provides a more active form of available chlorine which SWIFTLY destroys thermoduric and thermophilic organisms. Oakite Bactericide is completely soluble . . . drains freely . . . leaves no white residues on equipment. FREE booklet gives COMPLETE details. Write for your copy TODAY!

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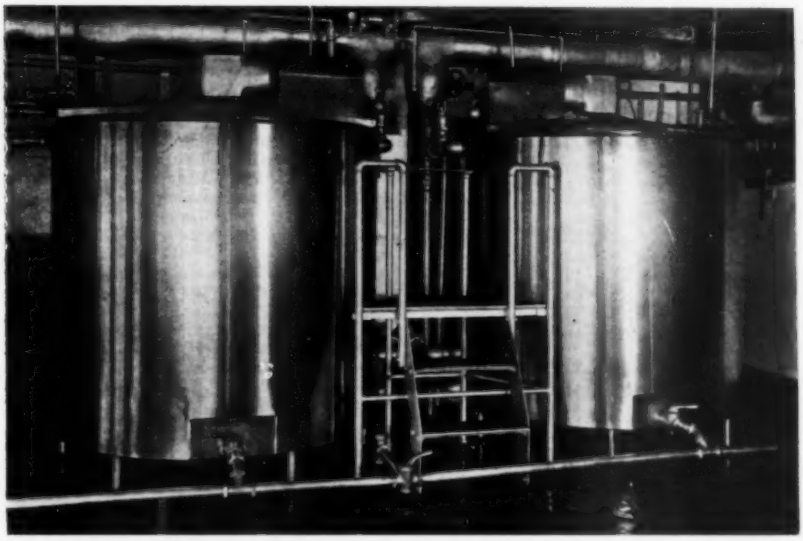
. . . for finest butter flavor and quality



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The American Dairy Science Association was organized to advance the general welfare of the dairy industry, especially by the improvement of dairy instruction by the stimulation of scientific research in all phases of the subject and by improvement in methods of conducting extension work.

Membership. Any person is eligible to membership who is formally announced by an Agricultural College or Experiment Station, or by the Bureau of Dairy Industry of the United States Department of Agriculture or by the Canadian Department of Agriculture as an instructor, extension worker, investigator, or administrative officer connected with the dairy industry, or any person filling a position of responsibility connected with the dairy industry who has had a college or university training in technical science, or any person filling a responsible position in the dairy industry of a professional character requiring a technical knowledge of dairying of a high order. The membership fee is $5.00.

The dues are $5.00 a year, $3.00 of which is for a year's subscription to the *Journal of Dairy Science.* Correspondence regarding membership and dues should be addressed to R. B. Stoltz, Ohio State University, Columbus, Ohio.

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RALSTON PURINA COMPANY SAINT LOUIS, MISSOURI

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We hope you will immediately write us saying you will accept and let us know when you will arrive in St. Louis. As soon as we know how many people will be here, during these two days, we will write you, giving further details of the program.

Sincerely

J. C. Thompson

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IT'S going to take healthy soldiers, healthy war workers, healthy families and healthy communities to win this war. That's the big reason why today it's more important than ever before to maintain highest standards in the purity of the milk they drink, despite the growing problems of changing conditions and changing personnel.

And SEAL-KAPS help to maintain high standards. For SEAL-KAPS not only tightly cover the pouring lip of the bottle from dairy to delivery but they *re*-cover the bottle after every use. Thus the milk is constantly guarded against the danger of contamination and pure milk is kept pure to the very last drop.

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SEAL-KAP COVERS THE DANGER ZONE

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IN DEMOCRATIC countries, police departments nab "bad actors" and keep them out of circulation. In food processing, we also encounter "bad actors"—elements and conditions that rob products of freshness, turn them rancid, destroy their flavor. But food technologists are already on their trail. They know, for example, that they must protect food products from irradiation by light . . . exclude oxygen or excessive humidity . . . guard against insect infestation.

Recent research shows that copper and iron are "bad actors." In the manufacture of shortening, they are eliminated to increase rancidity resistance of fats and oils. They affect stability of practically any food that contains fats or volatile flavors easily oxidized.

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But we have reduced the copper and iron in Diamond Crystal Salt until today they represent less than 1 part per million. Salt products in general, however, may contain significant amounts of both copper and iron.

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Why not check up on the salt you are now using? And if you would like help in keeping down the "bad actors," copper and iron, in your plant, write to Technical Director, Diamond Crystal, Dept. H-3, St. Clair, Michigan.

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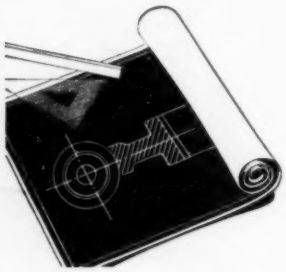

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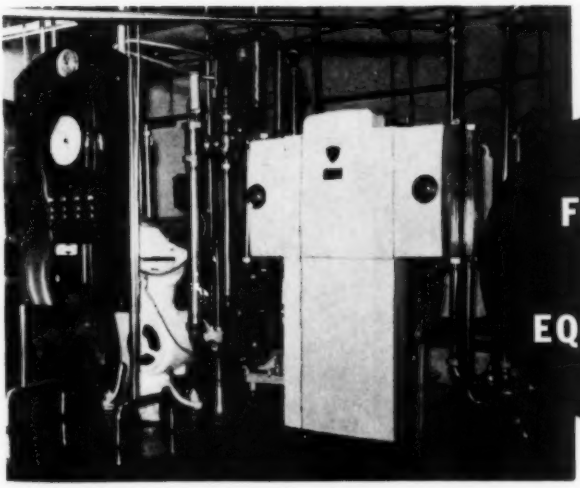

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Illustrated above: CP Full-Flo Plate HTST Pasteurizing Unit used on market milk at Franklin Cooperative Creamery Co., Minneapolis.

Hundreds of the finest dairy products plants in America and Canada have found the CP Full-Flo Plate Unit the most versatile and efficient equipment that money can buy. The "check list" at right will give you some idea of the variety of products being processed with this equipment.

Whatever your heat exchange problem, whether heating, cooling, preheating, regeneration or High Temperature Short Time pasteurizing, you'll find that CP Full-Flo Plate Equipment is tops. Ask for illustrated Bulletin E-5 which thoroughly explains the many exclusive CP advantages.

- ☐ **Heating and cooling cream.**
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DIFCO

Detection of
MOLDS *and* YEASTS
in Dairy Products

Bacto-Potato Dextrose Agar is recommended for determination of the mold and yeast count of butter. This product is also used extensively for isolation and cultivation of molds and yeasts in other dairy products.

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Bacto-Malt Agar is readily prepared and has a reaction of pH 5.5 after sterilization in the autoclave. This reaction may readily be adjusted to pH 3.5 by addition of lactic acid.

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In the Research and Development of Bacto-Peptone and Dehydrated Culture Media.

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